# A COMPARATIVE STUDY OF BONE MARROW ASPIRATION, IMPRINTS AND BIOPSY IN PANCYTOPENIA

**BY**

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KARNATAKA

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# A COMPARATIVE STUDY OF BONE MARROW ASPIRATION, IMPRINTS AND BIOPSY IN PANCYTOPENIA

**DOCTOR OF MEDICINE**

**in**

**PATHOLOGY**

# LIST OF ABBREVIATIONS

| | |
|---|---|
| ACD | Anemia of chronic disease |
| CRP | C - reactive protein |
| ESR | Erythrocyte sedimentation rate |
| MDS | Myelodysplastic syndrome |
| COPD | Chronic obstructive pulmonary disease |
| MEP | Megakaryocyte erythroid progenitor |
| CLP | Common lymphoid progenitor |
| BFU-E | Erythroid burst forming units |
| CFU-E | Erythroid colony forming unit |
| HSC | Hematopoietic Stem cell |
| CSF | Colony stimulating factor |
| BM | Bone marrow |
| TPO | Thrombopoietin |
| CFUMK | Megakaryocyte colony forming unit |
| DC | Dendritic cells |
| pDC | Plasmacytoid dentritic cells |
| MSC | Mesenchymal stem cells |
| Tf | Transferrin |
| PAS | Periodic acid Schiff |
| MPO | Myeloperoxidase |
| $Fe2^{+}$ | Ferrous ions |
| $Fe3^{+}$ | Ferric ions |
| TIBC | Total iron binding capacity |
| CMIA | Chemiluminescent microparticle immuno assay |

# ABSTRACT

**TITLE: A COMPARATIVE STUDY OF BONE MARROW ASPIRATION, IMPRINTS AND BIOPSY IN PANCYTOPENIA**

**BACKGROUND:** Pancytopenia is reduction in all the three major cellular elements of blood, hence it is the simultaneous presence of anemia, leucopenia and thrombocytopenia. It is not a disease entity but a triad of findings that may result from various disease processes, primarily or secondarily involving the bone marrow[1]. The complete hematological work up with good clinical correlation is of utmost importance to evaluate the cause of pancytopenia and planning further investigations. Bone marrow aspiration and biopsy are indispensable adjunct to the study of hematopoetic disorders.

**OBJECTIVE:** To correlate the bone marrow aspiration, imprint and bone marrow biopsy study findings in patients with pancytopenia.

**MATERIALS:** 64 patients of all age group requiring Bone marrow aspiration and biopsy diagnosis with pancytopenia referred to the hematology laboratory of Pathology department, Shri B.M. Patil Medical College, Hospital, and Research Centre during a period of 1st January 2020 to 31th March 2022.

**RESULTS:** Among 64 cases of pancytopenia studied, age of patients varying from 5-82 years, with higher incidence in age group 23-32 years showing male predominance.

Megaloblastic anemia was commonest cause with 47.7% followed by combined nutritional anemia 18.2%. Least common cause was myelofibrosis, multiple myeloma, MDS and Filariasis. Although the diagnostic accuracy of BMB was highest (100%) but diagnostic accuracy of BMI was also considerably high (85.9%) in comparison to BMA (84.4%) in diagnosing various pancytopenia cases. Current study showed a positive correlation with a p value of 0.03.

**CONCLUSION-**

In our study, one case of myelofibrosis was diagnosed on biopsy sections only. Hence, a finding of a dry tap should never be dismissed as being due to faulty technique and always need a bone marrow biopsy. The comparative evaluation of BMA, BMI and BMB is essential so that more rapid and efficient method may be defined for diagnosing various hematological disorders. The present study observed that although the diagnostic accuracy of BMB was highest (100%) but diagnostic accuracy of BMI was also considerably high (85.9%) in comparison to BMA (84.4%) in diagnosing various pancytopenia cases.

**KEY WORDS**: Bone marrow, Aspiration, Imprint, Biopsy, pancytopenia

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# INTRODUCTION

Pancytopenia is a hematologic condition characterized by a decrease in all three peripheral blood cell lines. It is characterized by the hemoglobin of less than 12 g/dL in women and 13 g/dL in men, platelets of less than 150,000 per mcL, and leukocytes of less than 4000 per ml (or absolute neutrophil count of less than 1800 per ml).[1,2] The etiology of pancytopenia is as varied as the presentation of the disorder. The causes ranges from megaloblastic anemia, aplastic anemia, leukemia, drug-induced, hypersplenism, infections like kala-azar, and systemic lupus erythematosus to name a few.[3] The identification of pancytopenia at various levels of diagnostic chains ranges from the outpatient departments to the fully sophisticated tertiary care centers. However, the final conclusive diagnosis needs a battery of investigations to be done.

The basic investigative modalities include the complete blood counts, peripheral smear, bone marrow examination brings the diagnosis up a notch by adding the definitive tag to the diagnosis. The utility of the history in terms of the medication use, recreational drug usage, intake of certain types of herbal medications, chemotherapy, and environmental exposure rules out many of the diagnostic dilemmas and narrows the diagnosis thereby reducing the need for extensive investigation.

Evaluation of the peripheral smear and bone marrow material forms the cornerstone in the diagnostic armamentarium. The peripheral pancytopenia is confirmed and further evaluated in the bone marrow evaluation providing an insight into the actual synthesis and creation of the cellular lineages.

However, on the contrary the disease progression might be slower and the patients have to be followed up very closely to document the diagnosis.[4]

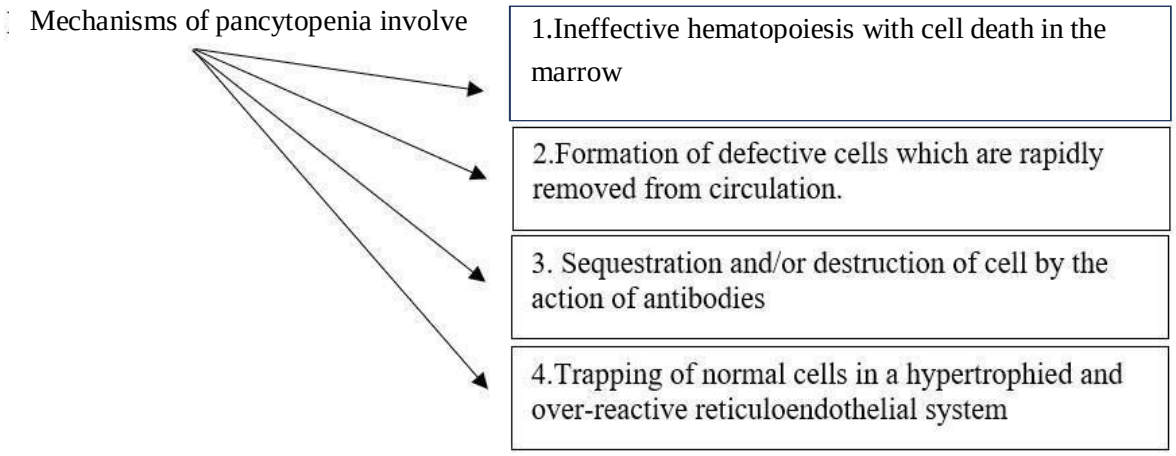
In this study, we have targeted the evaluation of pancytopenia using the comparative analysis of peripheral smear, bone marrow aspiration, imprint smears and bone marrow biopsy evaluation for identification of the etiology.

## OBJECTIVE

To compare bone marrow aspiration, imprint smears and biopsy in pancytopenia cases.

## REVIEW OF LITERATURE

Pancytopenia is not a disease entity but a triad of findings that may result from various disease processes, primarily or secondarily involving the bone marrow.[5] It is a striking feature of many serious and life-threatening illnesses, ranging from simple drug-induced bone marrow hypoplasia, megaloblastic anemia to fatal bone marrow aplasias and leukemias.[6]




Pancytopenia's underlying etiology differs depending on the geographical location. Due to the vast range of probable etiologies for pancytopenia, including nutrient deficiencies, infections, neoplastic entities, and bone marrow failure syndromes, diagnosis is time consuming and laborious.[7]

To determine the origin of pancytopenia and to plan future investigations and treatment, a thorough hematological workup that includes a good peripheral blood smear examination, bone marrow aspiration, imprint smear, and biopsy with clinical correlation is crucial.[8] Biopsy enables the investigation of the cellularity of the marrow, the detection of focal lesions, and the degree of infiltration by various pathological entities. Bone marrow aspirate is beneficial for

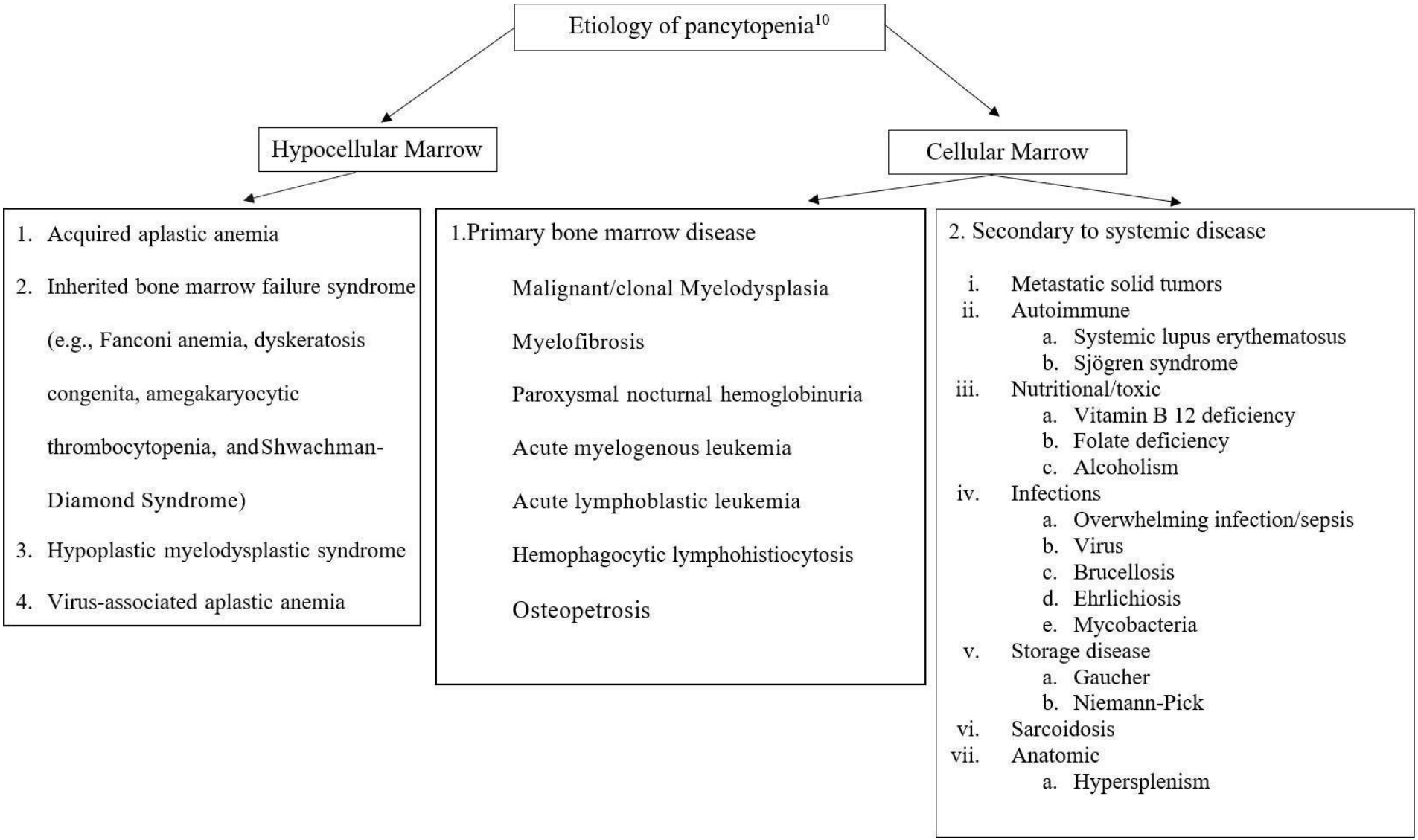
studying the cytological features.[9] Within the last three decades, the improved trephine design of the needle, the improvement in biopsy technique, and the technical progress in their preparations have provided additional impetus to the study of bone marrow as an organ with its architecture and components intact in their natural spatial context. This has offered a broader basis for comprehension of its function in health and disease.[4,5]

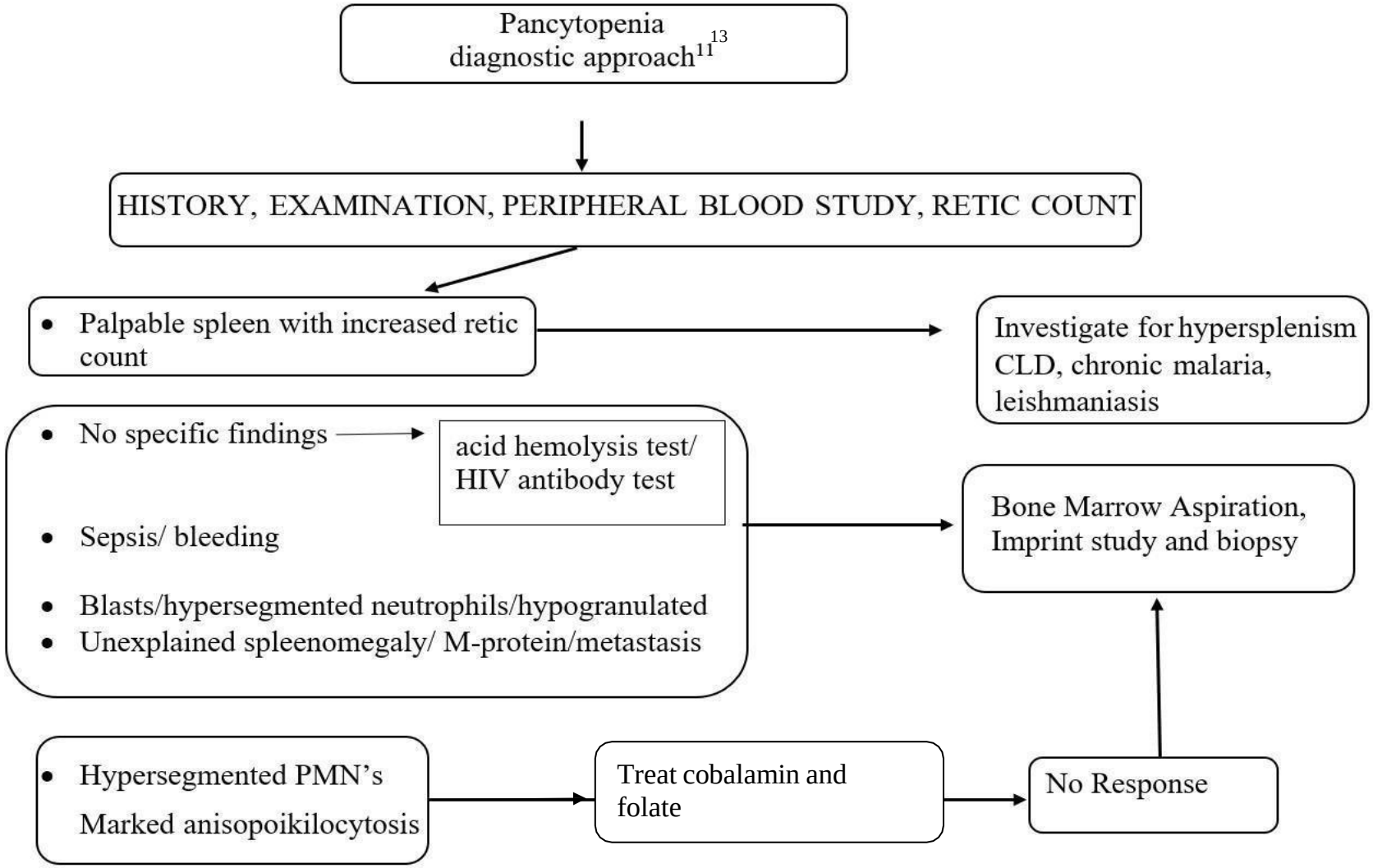
The biopsy is the gold standard for assessment of cellularity, pattern, the extent of tumor infiltration, and focal infiltration. It's more helpful in diagnosing granulomatous pathology and metastatic deposits of tumors eliciting a fibrotic response.[2]

Studies have compared the role of bone marrow aspirate cytology and trephine biopsy for diagnosing various cases of pancytopenia, but fewer studies have compared the relative value of imprint cytology with aspirate and trephine biopsy.[6]

Appropriately prepared imprint cytology smears do not only adequately provide the cellular composition of marrow but might also define the topographical architecture of marrow. Imprint cytology smears should therefore be standard practice for evaluating any marrow.[2]

A study done on 58 cases by Metikurkea SH et al.[11] stated that the familiar cause of pancytopenia was megaloblastic anemia. Both the procedures were complementary to one other and should be done at the same time along with imprint smears for total bone marrow workup and evaluation.[11] In an evaluation study done by Pathak R et al.[12] on 503 cases, the most common cause of pancytopenia was hypoplastic anemia followed by hematological malignancies, megaloblastic anemia, leishmaniasis, and Gaucher disease. Bone marrow examination was able to diagnose the cause of pancytopenia in 76.47% of the cases.[7]





## ROLE OF BONE MARROW STUDY

Mosler performed the first in vivo marrow procedure in 1876, harvesting marrow fragments from a leukemia patient using a standard wood drill. Studies conducted by Arinkin in 1929 proved marrow aspiration to be a safe, simple, and effective method fifty years later.[14]

## HEMATOPOIESIS:

For the orderly proliferation, differentiation, and circulation of blood cells, bone marrow offers a distinctive microenvironment.[15] Blood cell formation takes place at several anatomical places when a person develops from an embryo to an adult. It begins in the embryo's yolk sac and proceeds on to the liver and, to a lesser extent, the spleen. After birth, bone marrow is the only significant source of blood cell synthesis and begins to take over the role of hematopoiesis in the fourth month of fetal life. Significant structural and functional variations can be seen in these blood cells. However, the bulk of blood cells develop from a single hematopoietic stem cell (HSC), despite these distinctions. Hematopoiesis is the term used to describe the processes involved in the generation of various blood cells from HSCs.[15]

These HSCs have two distinct and crucial characteristics, namely pluripotency and the ability to self-renew, which are necessary for the ongoing maintenance of hematopoiesis. One HSC has the capacity to develop into all types of adult blood cells attributable to its pluripotency. One of the daughter cells of an HSC has the ability to replenish itself in order to prevent the depletion of stem cells.[15]

Several diverse early progenitor cell types with limited differentiation potential are produced by HSCs. In the end, these cells specialise to form lymphoid and myeloid cells. Colony forming

units (CFUs), which differentiate towards a specific lineage, are produced by early progenitor cells. These CFUs generate cell populations made up of particular mature cells. Precursors with morphological differentiation are now created, including myeloblasts, monoblasts, proerythroblasts, and megakaryoblasts. [16] Final differentiation of these cells results in the production of adult granulocytes, red blood cells, and platelets. [15]

Hematopoietic growth factors like erythropoietin, granulocyte- macrophage colony stimulating factor (GM- CSF), granulocyte colony stimulating factor (G- CSF), and thrombopoietin, which act through receptors which are expressed on the committed progenitor cells having restricted potential for differentiation, control the bone marrow's response to the fleeting physiological requirements.[16] The mature B cells, T cells, and Natural Killer (NK) cells are produced after the lymphoid committed precursors have differentiated into pre cells.(Fig 1)[16]

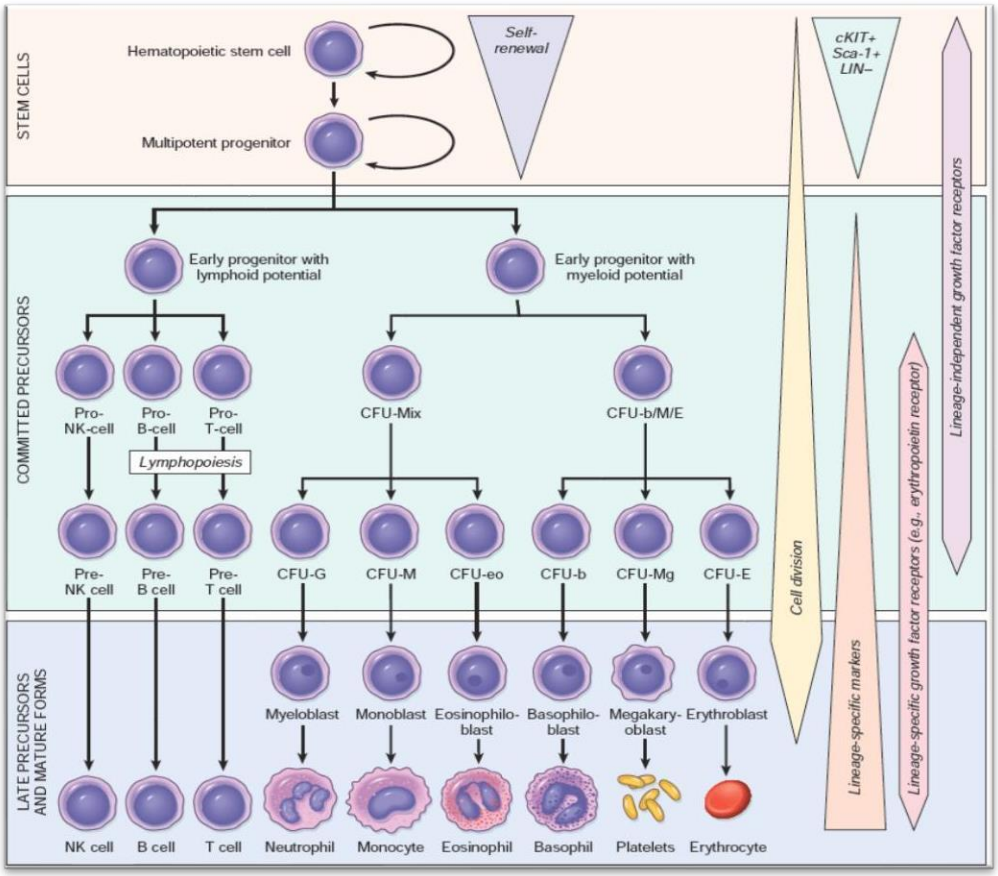



FIGURE 1: HEAMATOPOIESIS[16]

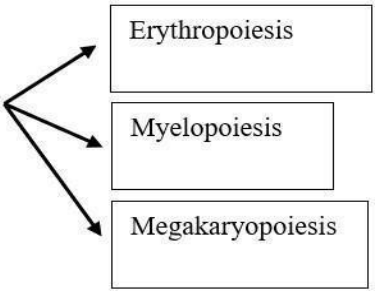
## CONSTITUENTS OF BONE MARROW

The major categorization of the bone marrow constituents is done into:

a. Hematopoietic cells and[16]

b. Stromal cells[16]

Hematopoiesis is further classified into:




## ERYTHROPOIESIS :

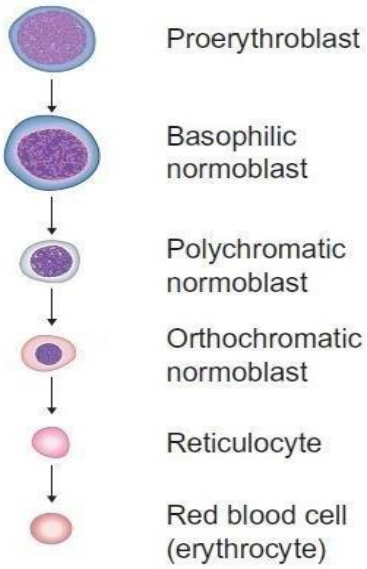



FIG 2A[17] NORMAL ERYTHROIPOIESIS

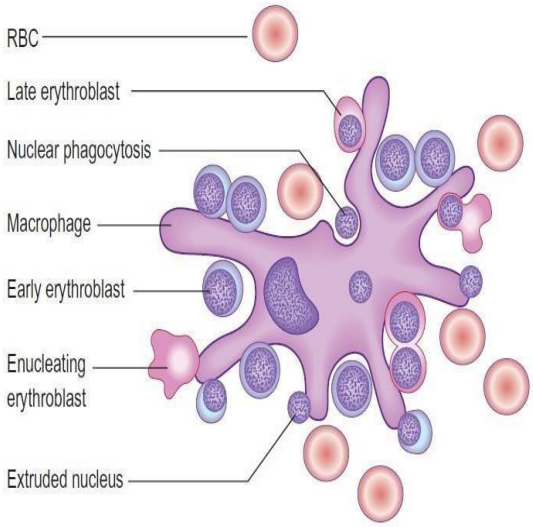



FIG2B[17] ERYTHROID ISLAND SHOWING NORMAL ERYTHROID PROGENITORS AND MATURE RBC'S.

1. All myeloid lineages are generated by the common myeloid progenitor (CMP) (i.e. granulocytic, erythroid, megakaryocytic). Following the CMP, megakaryocyte-erythroid progenitors (MEP) and granulocyte-macrophage progenitors (GMP) are produced (MEP). [17]
2. Natural killer (NK) cells and T- and B- lymphocytes are both derived from the common lymphoid progenitor (CLP). [17]
3. The most immature lineage-specific erythroid progenitor cells are the erythroid burst-forming units (BFU-E) and the most mature are the erythroid colony-forming units (CFU-E).[17]

   Proerythroblasts, the earliest morphologically recognizable BM red cell precursors, are formed from the CFU-E. Proerythroblasts move through various cytological groups that are determined by morphology.

   The basophilic erythroblasts, early and late polychromatic erythroblasts, and reticulocytes are these, in ascending order of increasing maturity. (Fig.2.a,b) [17] (Fig.4) [17]
4. The BM contains erythroid islands (Fig. 3)[17] that are made up of one or more central macrophages and one or two layers of erythroblasts.[17]

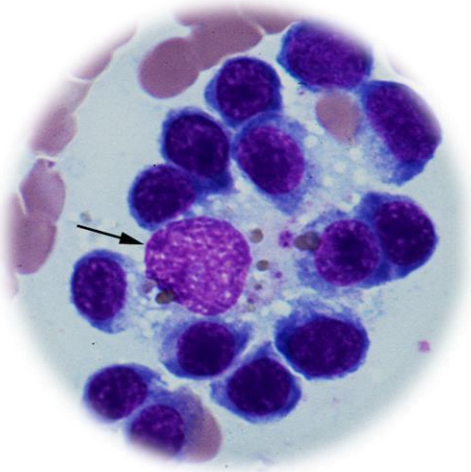


FIG.3[17]: ERYTHROID ISLANDS IN NORMAL BONE MARROW. ERYTHROID ISLANDS CONSISTING OF EARLY AND LATE POLYCHROMATIC ERYTHROBLASTS SURROUNDING MACROPHAGES

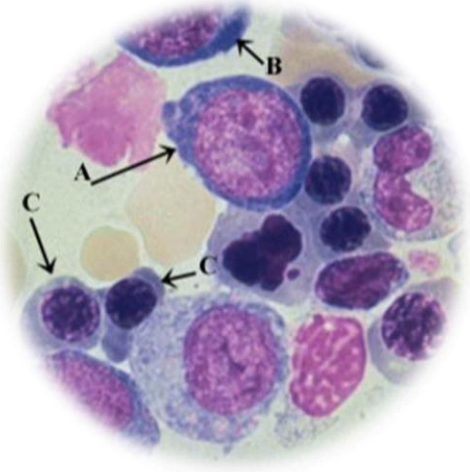



FIG.4[17]: (A) PROERYTHROBLAST (B) BASOPHILIC ERYTHROBLAST (C) POLYCHROMATOPHILIC ERYTHROBLAST

| NORMAL BONE MARROW ERYTHROID PRECURSORS | MORPHOLOGY |
|---|---|
| PROERYTHROBLAST (PRONORMOBLAST) (A) | >15μm cell with large central nucleus with open chromatin, bluish cytoplasm. |
| BASOPHILIC ERYTHROBLAST (BASOPHILIC NORMOBLAST) (B) | slightly smaller than the proerythroblast (10-15μm) with a central nucleus with chromatin that is starting to condense (clump/darker purple); cytoplasm dark blue (often more intensely blue than the proerythroblast). |
| POLYCHROMATOPHILIC ERYTHROBLAST (POLYCHROMATOPHILIC NORMOBLAST) (C) | smaller than the basophilic erythroblast (10-12μm) with a much smaller nuclear to cytoplasmic ratio; nucleus has condensed/clumped chromatin; cytoplasm now turning lighter/grayish/blue as hemoglobin is being produced. |

**GRANULOPOIESIS :**

1. Granulopoiesis is the process through which granulocytic cells (monocyte-macrophage series cells, neutrophils, eosinophils, and basophils) are produced within the BM.[17](Fig.5)[17]
2. It begins with the differentiation of the human stem cell into the common myeloid progenitor (CMP). The CMP continues to grow until it becomes the bipotent granulocyte-macrophage progenitor (GMP).[17]
3. The GMP develops into cells that are irreversible committed to maturing into granulocytic cells (CFU-G) or macrophages (CFU-M).[17]

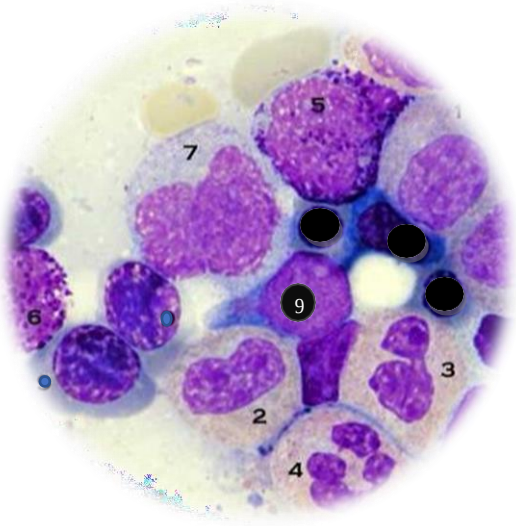



FIG 5: 1- MYELOCYTES, 2- METAMYELOCYTES, 3- BAND FORM, 4- SEGMENTED NEUTROPHIL, 5- BASOPHILIC MYELOCYTE, 6- MATURE BASOPHIL, 7- MONOCYTE, 8- SMALL LYMPHOCYTE, 9-MYELOBLAST

**MONOCYTOPOIESIS (THE MONONUCLEAR PHAGOCYTE SYSTEM) :**

1. The process by which tissue macrophages and peripheral blood monocytes are created is known as monocytopoiesis. The GMP, which serves as both the common ancestor for granulocytes and monocytes, is the source of the monocyte-macrophage lineage. The GMP passes through the monocyte maturation pathway under the control of GM-CSF, M-CSF, and IL-3, as well as up-regulation of the basic leucine zipper (bZIP) transcription factor MafB.[17]
2. In order of increasing maturity, monoblasts, promonocytes, and BM monocytes are the morphologically distinguishable antecedents of the monocyte series; only the first two of these go through division. After 20–40 hours, the blood monocytes cease to circulate and change into tissue macrophages.[17]
3. These can be found in the BM and other tissues. They play a part in erythropoiesis and the phagocytosis of cell debris.[17]
4. Normally, there is a steady loss of tissue macrophages (such as through alveolar

macrophage shedding), which is countered by the production of new macrophages from blood monocytes and to a lesser amount by the division of some existing macrophages. The mononuclear phagocyte system is the collective name for the group of cells responsible for producing macrophages.[17]

**LYMPHOPOIESIS:**

1. The HSC differentiates under the influence of IL-7 and FLT3 to produce the common lymphoid progenitor (CLP), a precursor of mature lymphocytes.(Fig.7)[17]
2. These produce all different types of lymphocytes, including B-cell, T-cell, and NK cell progenitors. Antigenic stimulation is not necessary for lymphopoiesis to take place in normal BM, which provides the body with mature B-lymphocytes or T-lymphoid progenitors that develop into T cells in the thymus.[17]
3. Mature B and T cells that have just developed enter the bloodstream and subsequently move to peripheral lymphoid tissues such the spleen, lymph nodes, Peyer's patches, and Waldeyer's ring.[17]

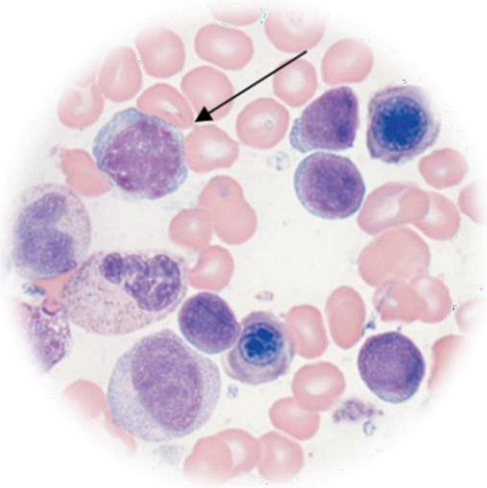


FIG.6[17]: NORMAL LYMPHOBLAST IN NORMAL PEDIATRIC MARROW. LYMPHOBLASTS ARE LARGER THAN LYMPHOCYTES WITH FINER, LESS CONDENSED CHROMATIN. (MAY– GRUNWALD–GIEMSA STAIN. 1000x)

**Plasma cells:**

Less than 1% of the typical BM's cells are plasma cells. They contain a deep basophilic cytoplasm, a pale perinuclear zone that corresponds to the location of the Golgi apparatus, cytoplasmic vacuolations, an eccentrically positioned nucleus, and a moderate quantity of condensed chromatin. They are 14–20 μm in diameter.[17](Fig.8)[17]

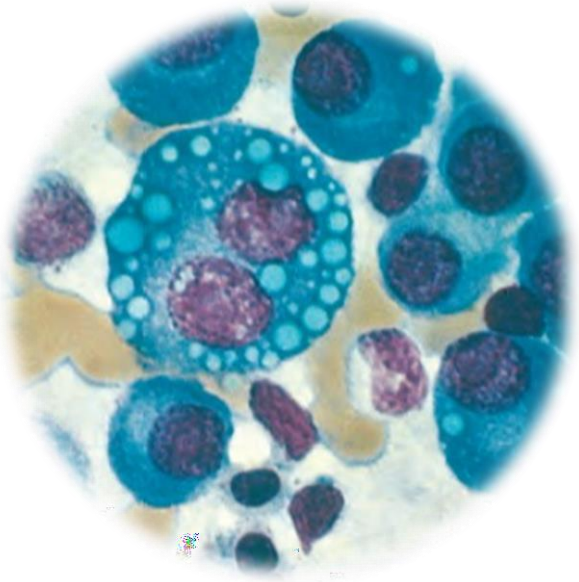


FIG 7[16]: PLASMA CELL WITH RUSSELL BODY.

**MEGAKARYOPOIESIS:**

The process by which megakaryocytes and platelets form in the bone marrow is known as megakaryopoiesis. Humans produce 1011 platelets each day, and when needed, output can rise 20-fold.[17]

Megakaryocytes are produced from the megakaryocyte erythroid progenitor (MEP) during a cascade of differentiation. Megakaryocyte colony-forming units(CFU-MK) are produced by the bipotent MEP under the control of thrombopoietin (TPO), the main regulator of platelet formation, IL-6, and IL-11. [17]

CFU-MK are a diploid cell population in which cell division follows DNA synthesis and nuclear division.[17]

Megakaryoblasts, the earliest morphologically discernible member of the megakaryocyte series, are formed as CFU-MK mature further. Romanowsky-stained megakaryocytic cells reveal four distinct kinds.(Fig.8)[17]

1. group I - Megakaryoblasts (megakaryocytes)
2. group II - Promegakaryocytes (megakaryocytes)
3. group III - Granular megakaryocytes which produce platelets. (Fig.9) [17]
4. 'Bare' nuclei

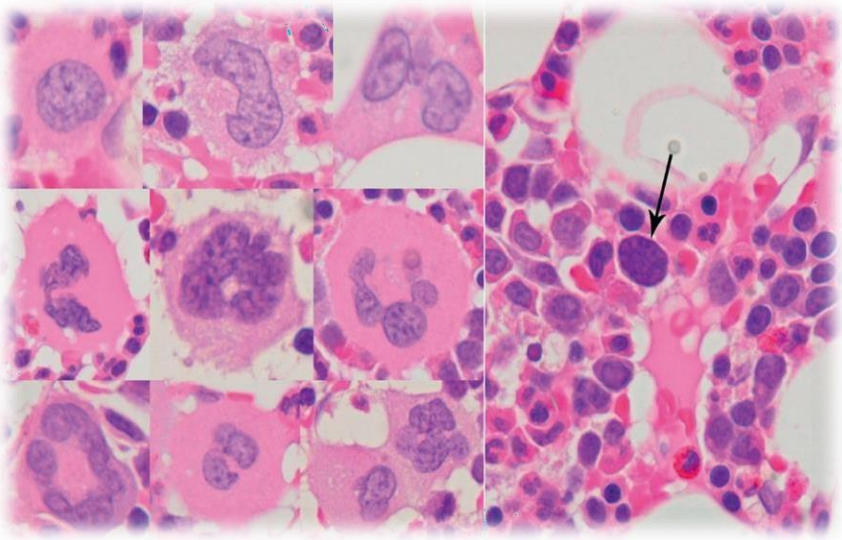


FIG.8[17]: THE THREE ROWS OF IMAGES ON THE LEFT SHOW THE VARIABLE MORPHOLOGY OF MEGAKARYOCYTES IN A SINGLE TREPHINE BIOPSY. A BARE NUCLEUS OF A SENESCENT MEGAKARYOCYTE IS ILLUSTRATED (ARROW)

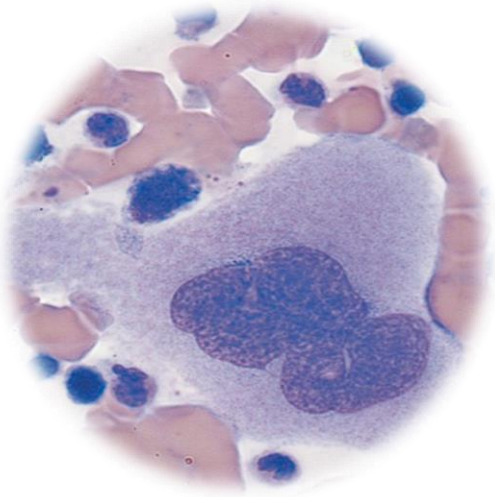


FIG.9[17]: GRANULATED MEGAKARYOCYTE

TABLE 1. NORMAL RANGE OF DIFFERENTIAL COUNTS ON ASPIRATED MARROW[10]

| OBSERVED RANGE (%) | | MEAN (%) |
|---|---|---|
| **NEUTROPHILIC SERIES (TOTAL)** | 49.2 – 65 | 53.6 |
| □ Myeloblasts | 0.2-1.5 | 0.9 |
| □ Promyelocytes | 2.1-4.1 | 3.3 |
| □ Myelocyte | 8.2-15.7 | 12.7 |
| □ Metamyelocyte | 9.6-24.6 | 15.9 |
| □ Band form | 9.5-15.3 | 12.4 |
| □ Segmented | 6.0-12.0 | 7.4 |
| **EOSINPHILIC SERIES (TOTAL)** | 1.2-5.3 | 3.1 |
| □ Myelocyte | 0.2-1.3 | 0.8 |
| □ Metamyelocyte | 0.4-2.2 | 1.2 |
| □ Band form | 0.2-2.4 | 0.9 |
| □ Segmented | 0-1.3 | 0.5 |
| **BASOPHIL AND MAST CELLS** | 0-0.2 | <0.1 |
| **ERYTHROID SERIES (TOTAL)** | 18.4-33.8 | 25.6 |
| □ Pronormoblast | 0.2-1.3 | 0.6 |
| □ Basophilic | 0.5-2.4 | 1.4 |
| □ Polychromatophilic | 17.9-29.2 | 21.6 |
| □ Orthochromatic | 0.4-4.6 | 2.0 |
| **LYMPHOCYTE** | 11.1-23.2 | 16.2 |
| **PLASMA CELL** | 0.4-3.9 | 1.3 |
| **MONOCYTE** | 0-0.8 | 0.3 |
| **MEGAKARYOCYTE** | 0-0.4 | <0.1 |
| **RETICULIN CELLS** | 0-0.9 | 0.3 |
| **MYELOID TO ERYTHROID RATIO** | 1.5-3.3 | 2.3 |

## BONE MARROW ARCHITECTURE - NORMAL ANATOMY

The aspirate and biopsy of bone marrow allow for the evaluation of bone marrow architecture and the distribution of biological components, such as bone and stromal cells. A zone of cartilage or cortical bone follows the collagenous periosteal connective tissue that makes up the biopsy's outermost components (depending on the age of the patient). Following this, the bone separates into a network of trabeculae, with intertrabecular voids between them. (Fig.10)[17]

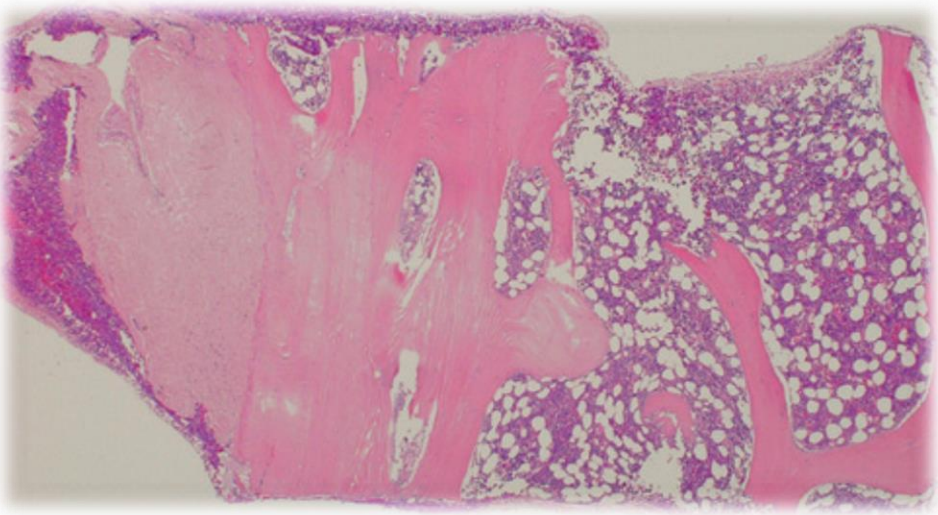


FIG.10[17]**:** LOW POWER VIEW OF A BONE MARROW BIOPSY. PERIOSTEAL CONNECTIVE TISSUE IS SEEN EXTERNAL TO CORTICAL BONE.

Fat cells, stromal cells, histiocytes, extracellular matrix, and blood vessels nourish the hematopoietic cells that are present in these intertrabecular gaps.[17] Three zones that each contain a different type of hemopoietic cell can be found within the intertrabecular sections. (Fig.11) [17]

1. Immediately surrounding the trabecular bone, the endosteal or paratrabecular zone is primarily made up of myeloid progenitor cells.
2. Erythroid colonies and myeloid cells that are maturing are found in the intermediate zone.
3. Aging adipose tissue and fat globules are found in the central zone of the intertrabecular space. Sinusoids and megakaryocytes are also present, along with erythroid cells and

myeloid cells that are developing. In the middle and central zones, small arteries and arterioles are frequently observed; these structures may have cuffs of immature myeloid cells surrounding them.[17]

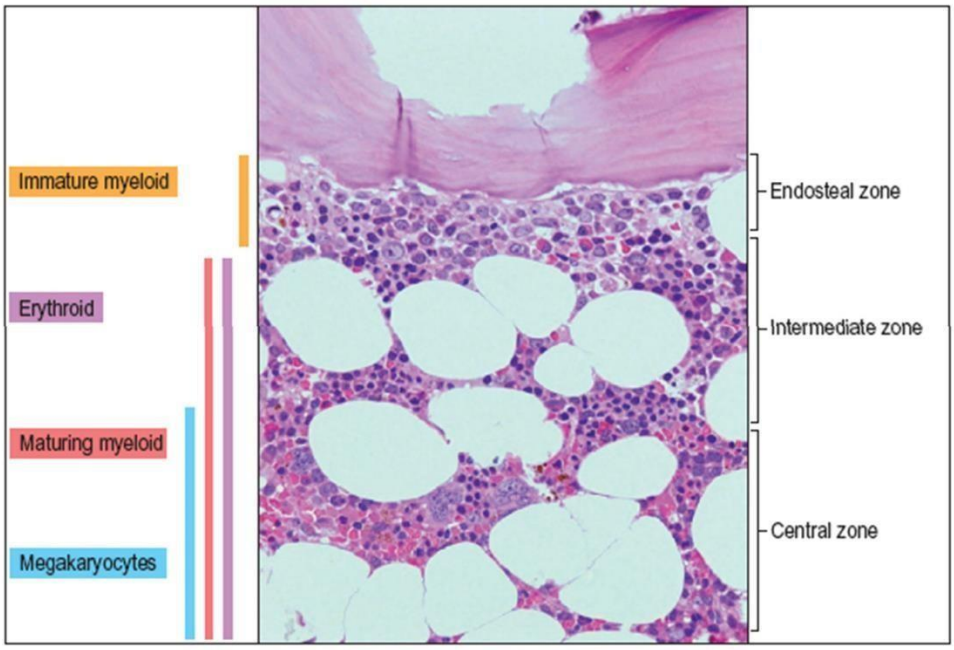



FIG. 11[17]: ORGANIZATION OF THE BONE MARROW: ZONES AND DISTRIBUTION OF VARIOUS CELL TYPES. × 100

TABLE 2.[10] CELLULARITY VARIES FOR DIFFERENT AGES

| Age | Cellularity |
|---|---|
| Newborn to 3 months | 80–100% |
| Childhood | 60–80% |
| 20–40 years | 60–70% |
| 40–70 years | 40–50% |
| >70 years | 30–40% |

TABLE 3: INDICATION FOR BONE MARROW ASPIRATION WITH OR WITHOUT BIOPSY[18]

| | |
|---|---|
| 1 | Unexplained anemia |
| 2 | Unexplained macrocytosis |
| 3 | Unexplained microcytosis |
| 4 | Unexplained thrombocytopenia |
| 5 | Pancytopenia |
| 6 | Leukoerythroblastic blood smear and suspected bone marrow infiltration |
| 7 | Suspected acute leukemia |
| 8 | Assessment of remission status after treatment of acute leukemia |
| 9 | Suspected myelodysplastic syndrome or myelodysplastic/myeloproliferative disorder |
| 10 | Suspected chronic lymphocytic leukemia and other leukemic lymphoproliferative disorders |
| 11 | Suspected non-Hodgkin lymphoma |
| 12 | Suspected hairy cell leukemia |
| 13 | Staging of non-Hodgkin lymphoma |
| 14 | Suspected multiple myeloma or other plasma cell dyscrasia |
| 15 | Suspected storage disease |
| 16 | Fever of unknown origin |
| 17 | Confirmation of normal bone marrow if bone marrow is being aspirated for allogeneic transplantation |

CONTRAINDICATIONS[18]

Contraindications to bone marrow aspiration/biopsy include.

| **Absolute contraindication** | |
|---|---|
| 1 | Hemophilia and other major disorders of coagulation. |

| **Relative contraindication** | |
|---|---|
| 1 | Skin infection |
| 2 | Previous radiation therapy in the area of sampling and an uncooperative patient. |

## PROCEDURE FOR ASPIRATION AND PREPARATION OF SLIDES

### Patient Consent[18]

It is crucial that a consent form is signed and that an explanation is given before any bone marrow aspiration procedures are carried out.

### STERNAL PUNCTURE[14]

The method of bone marrow aspiration from the sternum was first employed in 1929 by Russian doctor Mikhail Innokent'evich Arinkin, who utilised a lumbar puncture (spinal tap) needle to take a sample from this location. Since that time, sternal puncture has grown in popularity as one of the most frequent intraosseous diagnostic techniques used in haematology. Even though the posterior ilium in adult humans produces the most bone marrow, the sternum is still one of the most often used sites to take aspirates for haematological diagnosis.

This preferential selection might be brought on by the locus of hematopoietic marrow's proximity to the body's surface and accessibility.

### ILIAC PUNCTURE[14]

Another location from which hematopoietic marrow can be extracted is the ilium. The ilium can be easily pierced in several locations; the crest, however, presents too much resistance to be a useful location for routine penetration, with the possible exception of newborns and young children whose compact bone is less dense. Aspirating bone typically takes place near the anterior superior iliac spine (anterior iliac puncture) or the posterior superior iliac spine (posterior iliac puncture). It should be noted that the ileum provides more resistance to penetration than the sternum, necessitating the use of heavier needles for this technique.

INSTRUMENTATION

Bone marrow aspirate samples can be taken from the anterior iliac crest using the Salah, Klima, and Islam sternal puncture needles. However, the sternum, a much softer bone, is where the Salah and Klima needles were initially intended to be used. They may be weaker and less suited for puncturing the harder ilium, where the risk to the needle may be greater. Islam sternal puncture needles are significantly more robust, and since they have a T-bar and dome handle, they are simpler to hold and control when puncturing a hard bone. The Islam needle might be the needle best suited for anterior iliac puncture, depending on the operator's preferences.

Posterior Superior Iliac Puncture [14]

The biggest volume of hematopoietic (red) marrow is located in the area of the posterior superior iliac spine, which is the thickest part of the ilium in both children and adults.

Large amounts of red marrow can be aspirated with ease, and it is readily accessible. Since this ilium segment is far from any important structures, difficulties are unlikely. Additionally, the patient is unable to watch the process, preventing the anxiety that is frequently linked to sternal punctures. The cortical bone in this area is also noticeably less dense than it is in the area of the anterior iliac spine, making it easier to penetrate. This is an added benefit.

Instrumentation

In order to aspirate bone marrow from the posterior (as well as the anterior) iliac region, the Salah and Klima needles have been frequently utilised. They are not well suited for iliac puncture since, as previously mentioned, they were originally created to retrieve sternal samples of marrow.

Here, the configuration of the bone is different and the cortex thickness is higher. The bone marrow aspirate samples from the posterior iliac crests can only be collected using the Islam

posterior iliac puncture needles.

The Standard size steel instrument[2] consists of two parts, (a) The needle is 80 mm in total length, 2.35 mm in external diameter, and 1.78 mm in internal diameter, with the exception of the 1.25 mm distal end, which is bevelled at an 18° angle.

The needle's proximal end has been fitted with a broad metal bar designed for a solid grip and a female luer lock that can fit the male luer lock on a syringe's nozzle.

(b) The stilette has a solid 1.62 mm-diameter shaft, but its distal end has a 3.0 mm-long, three-faceted, sharp-pointed cutting tip that extends past the needle's distal end to allow for simple penetration of the soft tissue and bony cortex.

The male luer lock on the dome handle's interior has been placed into the proximal end of the stilette so that it fits the female luer lock on the needle. A hemispherical smooth dome-shaped solid nylon handle (c) measuring 30 mm in diameter and 15 mm deep with a softly machined edge of 5 mm caps off the proximal end of the stilette. If and when powerful thrusting is required, it will remain comfortably in the operator's palm because to its ergonomic design. Bone marrow aspiration for Posterior superior iliac spine is illustrated on**Fig.13**[14]

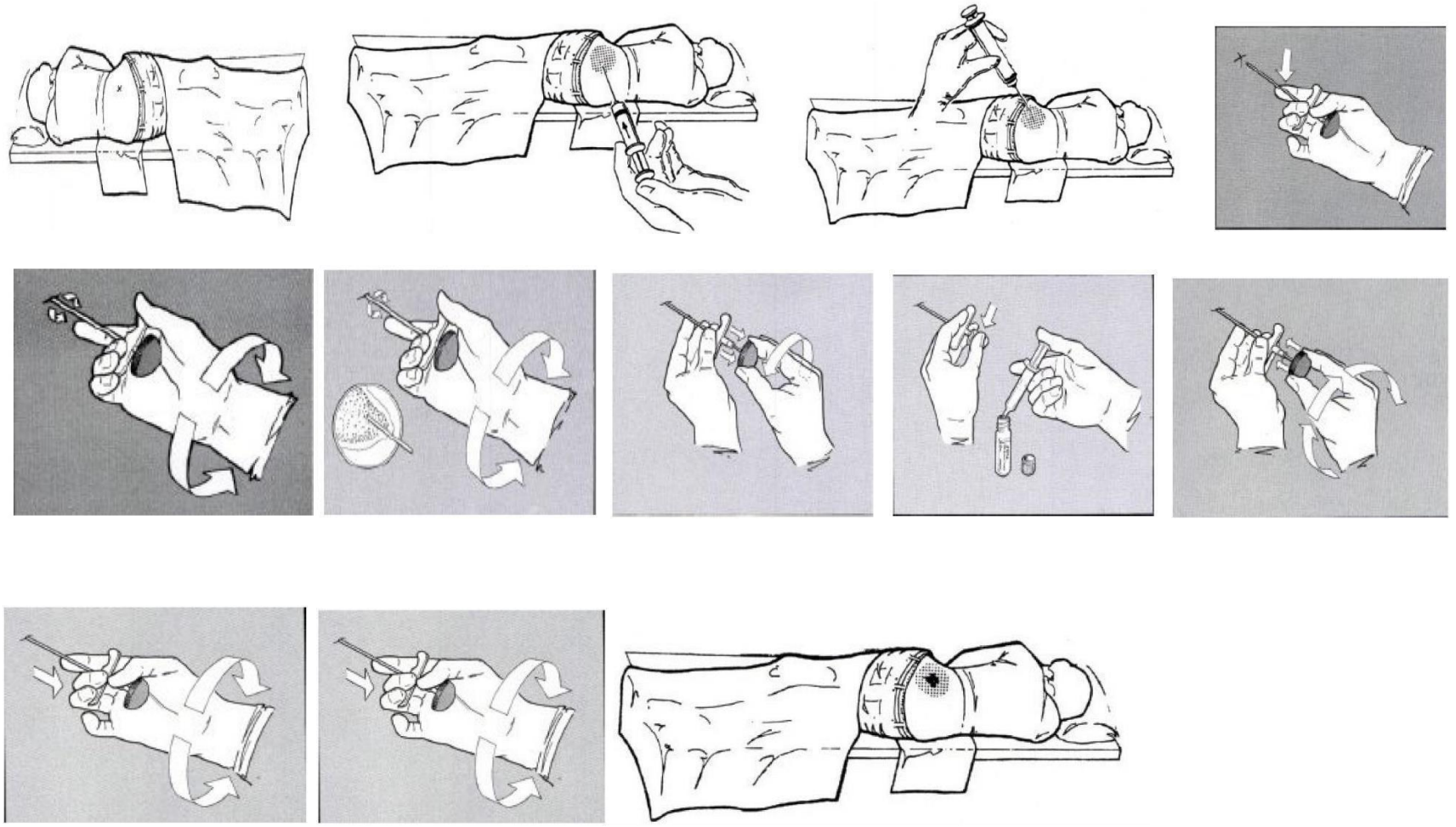


Fig12[14]**:** Aspiration from Posterior Superior iliac Spine

**PUNCTURE OF THE TIBIA**

Throughout the whole childhood, the tibia's proximal (epiphyseal) end has active marrow. At the age of 10, children's tibias have been successfully used to aspirate hematopoietic marrow. In all children under the age of two, it is standard procedure to choose the tibia as the preferred location of puncture. **Fig.13**

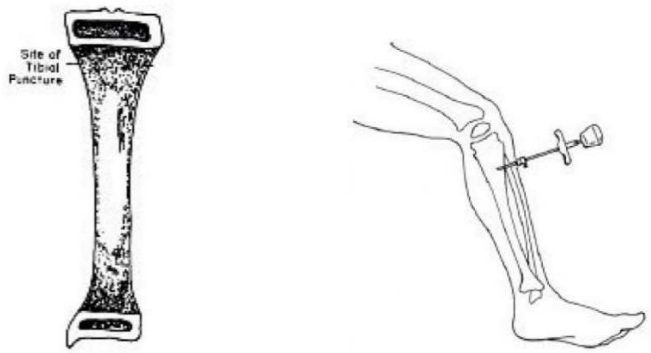



Fig.13[14] Aspiration from Tibia

Instrumentation

The tibia can be pierced using any of the available sternal puncture needles. You can use Salah, Klima, or needles that are comparable. Due to its enhanced design and simplicity of usage, the Islam sternal puncture needles are perfect for this application.

**QUANTITATIVE REQUIREMENTS FOR ASPIRATION AND SECTIONED TISSUE SAMPLING**[14]

The quantity of bone marrow that must be aspirated is small for standard bone marrow aspiration. Typically, 0.3 to 0.5 ml is enough to prepare a number of dry film smears.

Small quantities are also considered as desirable because they prevent hematopoietic cells from becoming diluted by circulating blood and its biological components.

When cytogenetic and/or fluorescent flow cytometric analyses are performed, larger quantities may be necessary.

To ensure the recovery of sufficient, histologically representative, undisturbed specimens, a 15-20 millimeter-long core of marrow tissue should be retrieved for solid tissue (needle) biopsies.

Excess bone marrow units from an aspirate can occasionally be collected, allowed to accumulate in a plasma/thromboplastin clot, and then subjected to the embedding and sectioning process to generate sections of variable, limited value.

## THE MULTIPORT ASPIRATION NEEDLE[14]

When diagnosing malignant and benign, hematologic and non-hematologic disorders, the technique of examining bone narrow aspirates taken through traditional marrow aspiration needles is frequently used. The fact that marrow fragments are only extracted from a small amount of tissue close to the open front end (mouth) of such needles is inherent in their form, although being almost completely underappreciated. Numerous marrow aspiration experiments in chosen people have shown that bone marrow displays patchy, random involvement in various disease conditions.

Standard aspiration needles only have one aperture for marrow access and sample. Cells close to or far from the point of aspiration may or may not have been included in their sample.

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Standard aspiration needles only have one aperture for marrow access and sample. Cells close

to or far from the point of aspiration may or may not have been included in their sample. A multipore aspiration needle has been created to help reduce the risk associated with collecting cells from several, distant loci of pathology. The probability of failing to sample tiny focal infiltrates in illnesses including lymphoma, leukaemia, preleukemia (MDS), and metastatic cancer is decreased by using this equipment (**Fig.15**)[14].

Experience has also shown that because to the superior depth of penetration provided by the posterior iliac crest, this needle is particularly helpful in posterior ilial (posterior iliac crest) aspirations.

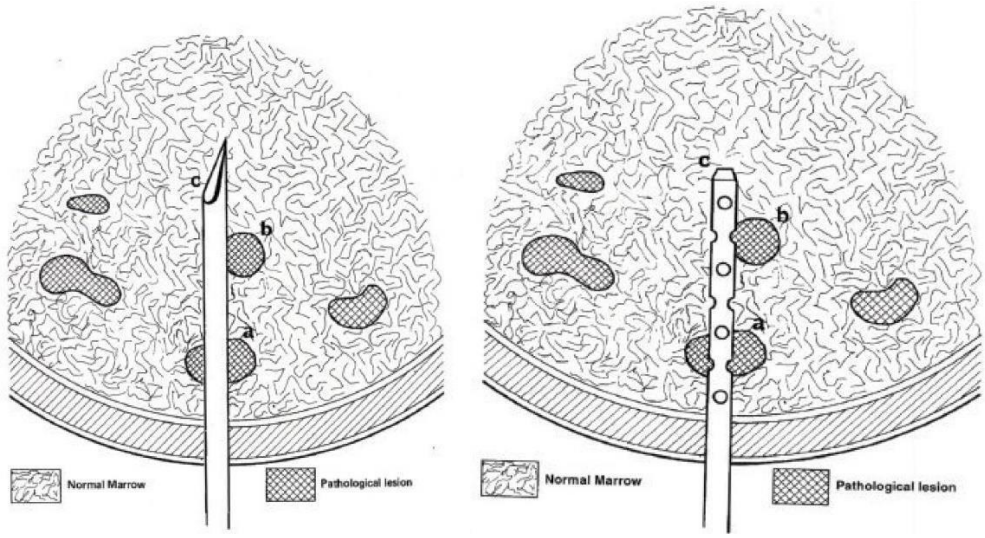



**Fig.14**[14]**:** Illustrating conventional needle miss pathological lesion whereas multiport aspiration needle exposed to wider areas of bone marrow.

Instrumentation

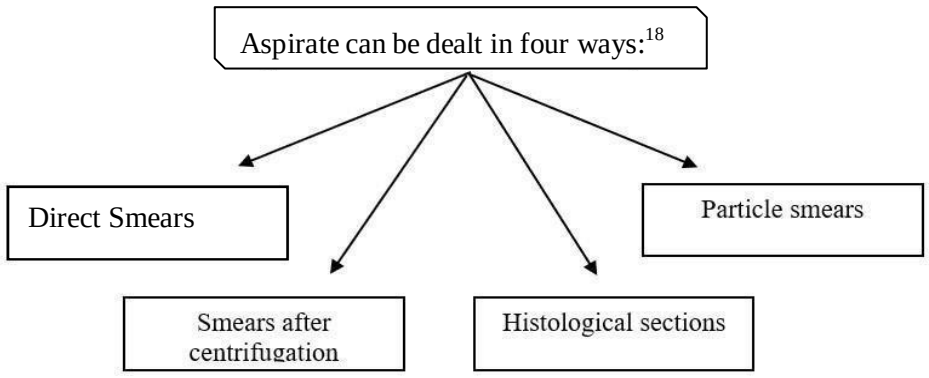
This instrument's structure and design are identical to those of the Islam posterior iliac puncture needle, but in addition to the one frontal aperture, it contains numerous side holes.

14 lateral ports, 4 rows of 8 holes in one plane, and 3 rows of 6 holes in another may be found on the 17.5 mm distal part of the needle. To prevent any weaknesses that would show if drilling for two ports were done on the same transverse plane, the rows of apertures are 2.5 mm apart and spiral-drilled. Each hole in the first row is about 1.0 mm in diameter and is drilled 3.0 mm from the end of the beveled point.

Aspiration Procedure

With this needle, you puncture the posterior superior iliac spine in the same way you would with any other needle. However, in this instance, care must be taken to ensure that the full perforated portion of the needle is inserted into the medullary cavity and away from the cortical entrance site.

By doing this, the possibility of aspirating air and soft tissue stuck to the ilium's externum is avoided.




**Aspirations films are suited for:**[18]

- Wedge spread films, films of crushed marrow fragments, study of cell markers, cytogenetic study, ultrastructural examination, culture of microorganisms, culture of hemopoietic precursors, and histological sections of fragments

**Complications of both aspiration and biopsy**[18]

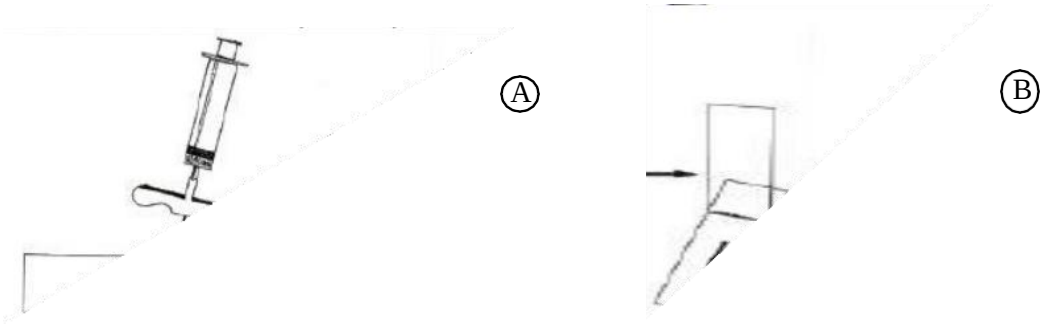
- Cardiac and great vessel laceration in case of sternal puncture
- Hemorrhage- if hemostatic defect is suspected, apply firm pressure
- Hemorrhage- also in abnormal vascularity of the bone as in Paget's disease
- Pneumothorax
- Sternomanubrial separation
- Breakage of the needle in patients with osteosclerosis.

**PROCESSING OF SPECIMENS**[14]

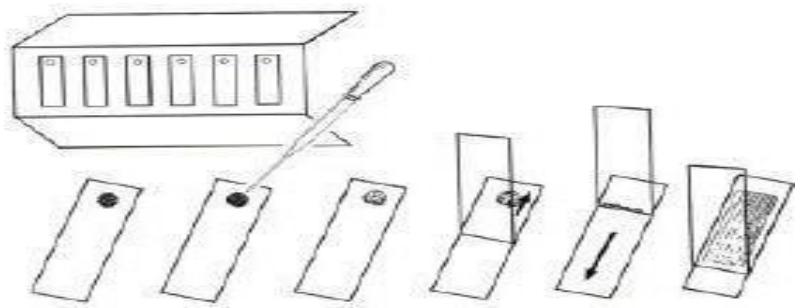
Aspirate Smear Preparation

As soon as the marrow has been aspirated, smears should be prepared. There are various methods for doing this.

(1) Typically, a sizable drop of marrow with adequate pieces can be applied straight on a slide from the aspirating needle. (A) or from the syringe upon its release from the needle (B). The marrow from the slide above is repeatedly collected, and dry film smears are prepared using the spreader slide's edge.




(2) A single drop of bone marrow can also be applied to numerous slides, the excess blood (but not the pieces) sucked off using a thin Pasteur pipette, and the remaining samples were then used to create smears.



**METHODS OF STAINING SMEARS**[18]

Cell preparations and imprints should be dried quickly by air, fixed for three to ten minutes with methanol or May-Grünwald alone, and stained twice: Following 3-5 minutes of 10 percent buffered Giemsa solution, 10-30 minutes of 50 percent May-Grünwald in buffer pH 6.8, and 1-3 minutes of running water. RBCs must be pink-orange (acidophilic) or buff-colored, not green or blue, at the right pH circumstances. The cytoplasm of leukocytes must be nearly translucent and have distinct granules. Staining can vary slightly, though, and testing is advised for automated techniques (slide stainers), which continue to be the gold standard for repeatability. Although MGG stain continues to be the standard stain, cell samples can also be quickly assessed using Diff-Quik(®) stain.[18]

Leishman stain

Flood the slide with the stain. After 2 min, add double the volume of water and stain the film for 30-45 min. Then wash it in a stream of buffered water until it has acquired a pinkish tinge (up to 2 min). After the back of the slide has been wiped clean, set it up right to dry.[18]

**Assessing and reporting bone marrow films** [14]

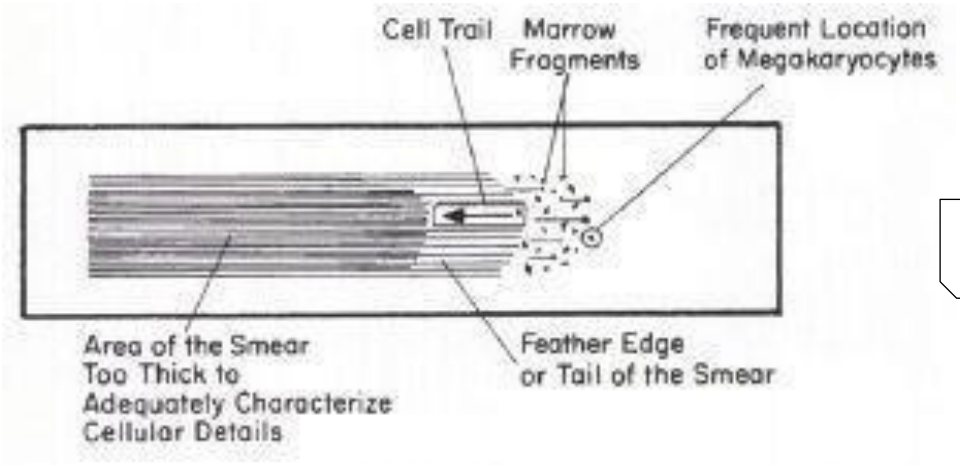



Fig.15 Cell trails

An evaluation of the marrow particles to determine the cellularity of the marrow should come first when reporting a bone marrow sample. By carefully examining multiple marrow pieces in one or more dyed films, the degree of cellularity can be classified as increased, normal, or decreased within broad boundaries. When the volumetric ratio of fat to hematopoietic cells in the bone marrow is roughly 50/50, the marrow is said to be normal. Age has an impact on the marrow's cellularity, so it's important to consider this physiological variation when evaluating the tissue. In comparison to children, adult bone marrow occupies a comparatively smaller percentage of the marrow cavity, and the ratio of fat cells to active marrow is higher. The marrow tends to continue to grow more fatty in persons 60 years of age or older; this is especially true for the manubrium sterni. It is well known that pregnancy causes a mild to moderate hyperplasia of the bone marrow.

The evaluation of the bone marrow should focus on the qualitative and quantitative characteristics of erythropoiesis, granulopoiesis, thrombopoiesis, and the presence or absence of any abnormal cells after assessing the cellularity.

The best place to accomplish this is in the cell trails. Near the fragments, megakaryocytes are most readily apparent. The existence or absence of ring sideroblasts should then be determined, together with the amount of iron present. The reviewer's general opinion of the material should be mentioned in conclusion.

An Effective, Useful Bone Marrow Report Should Comment on the Following[18]

1. Site: Under local anaesthetic, a bone marrow aspirate is taken from the sternum, the right or left posterior iliac crest, etc.
2. Cellularity in general: normal, elevated, or lowered. Prefixes can be used to further categorise the increase or decrease as mild, moderate, or marked. If there are few or no pieces in the aspirate, it will be impossible to evaluate the bone marrow's cellularity.
3. Erythropoiesis: normal, increased, or decreased activity; megaloblastic/ micronormoblastic /normoblastic maturation sequence; cytologic anomalies such as dyserythropoietic alterations; etc.
4. Granulopoiesis: activity (normal, enhanced, or decreased); maturation sequence; cytologic anomalies such as dysgranulopoiesis (giant metamyelocytes, Pelger-Huet forms, etc.); presence and number of myeloblasts and promyelocytes.
5. Megakaryopoiesis: numerical evaluation (increased, normal, or lowered); platelet production sequence; pleomorphism; and cytologic anomalies like dysmegakaryopoiesis (multinucleated forms etc.).
6. Plasma cell series: numerical evaluation (in percentage form), maturation, inclusion bodies, any anomalies, or malignant alterations.

7. Lymphocyte series: Malignancies, lymphoid aggregates, germinal centres, numerical evaluation (expressed in percentage), morphologic anomalies (big, tiny, cleaved, character of nucleoli), and numerical assessment (expressed in percentage).
8. Eosinophils and basophils: numerical assessment (expressed in percentage), any abnormalities.
9. Iron stores: content (normal, increased or decreased), the presence or absence of ring sideroblasts.
10. Miscellaneous: presence of macrophages, mast cells, granulomas, amyloidosis, gelatinous transformation of fat, pathologic lesions of the bone, osteoblasts, osteoclasts and metastatic neoplasms etc.
11. Differential cell count: The normal ranges for differential counts on aspirated bone marrow are given in Table 1.
12. Comment/conclusion: general opinion of the marrow's normality or abnormality If abnormal, specify the anomaly and any additional tests that should be carried out, such as vitamin B12 and/or folic acid assays for megaloblastic anemias, serum iron, TIBC, and other tests for iron deficiency anaemia, as well as any other studies that are suggested.

Preparation : Satisfactory /Adequate/ Inadequate

Cellularity : Normocellular/ Hypocellular / Hypercellular

Erythropoiesis : Normoblastic /Megaloblastic/ Micronormoblastic.

Myelopoiesis : Morphology/ Number/ Maturation.

M:E ratio : Normal / Reversed.

Lymphocytes : Morphology/ Number/

Megakaryocytes : Normal/ Hypolobulated/ Hyperlobulated. Plasma cells: Present/ Absent/ Number.

Abnormal cells : If any.

Iron Stores : Grade of iron stores according to Gales method.

Impression :

**Assessment of Iron stores in bone marrow**

Prussian blue method for estimating the amount of iron stored in bone marrow was developed by Perl in 1867. [19]

Principle: According to Perl, soluble ferrocyanide in the stain reacts with ferric iron deposits in tissue to generate insoluble Prussian blue dye (a complex hydrated ferric ferrocyanide compound) in situ. Ferric iron deposits are primarily found as ferric iron within the storage protein ferritin. [19] When viewed under a microscope, they appear as blue or purple deposits inside of cells.

Up to 30% of polychromatic erythroblasts have one tiny blue-black granule, which can be seen using Perls' acid ferrocyanide stain.

Sideroblasts are erythroblasts that have iron-containing siderotic granules that are randomly scattered throughout the cytoplasm.[19]

When there are five or more perinuclear siderotic granules, they are referred to be ring sideroblasts.

**Table 4: Grading for iron storage[19] (Figs.:16- 28)[20]**

| | |
|---|---|
| **0** | No stainable iron. |
| **1** | Small iron particles just visible in reticulum cell in oil immersion objective. |
| **2** | Small, sparse iron particle in low power field. |
| **3** | Numerous small granules in all marrow particles. |
| **4** | Large granules in small clumps. |
| **5** | Dense large clumps of granules. |
| **6** | Very large deposits obscuring marrow details. |

**Interpretation of iron stores:** [9, 20]

Minimum seven particles are required to see for the depletion of iron stores:

They are interpreted as:

| | |
|---|---|
| 0: | Absent iron stores |
| 1,2 | Normal |
| 3 | Slightly increased |
| 4 | Moderately increased |
| 5,6 | Markedly increased |

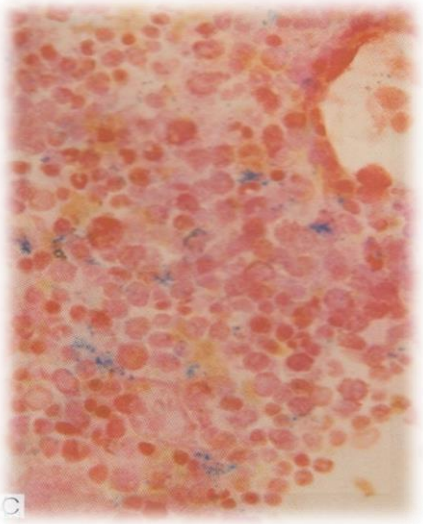


Fig.16[20]: Grade 3 iron stores

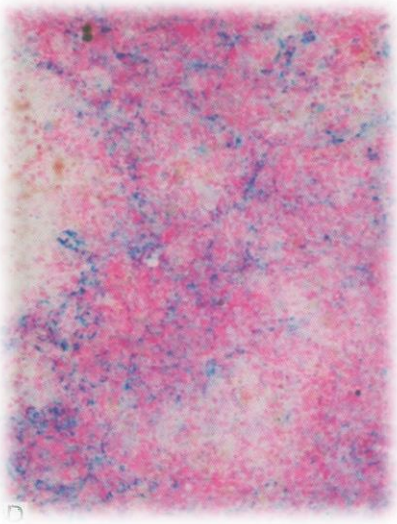


Fig.17[20]: Grade 4 iron stores

Fig.18[20]: Grade 5 iron stores

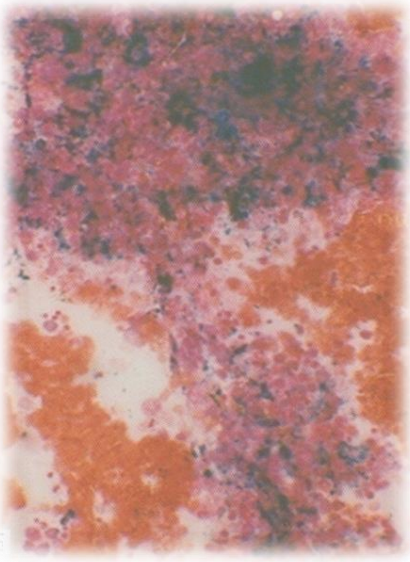
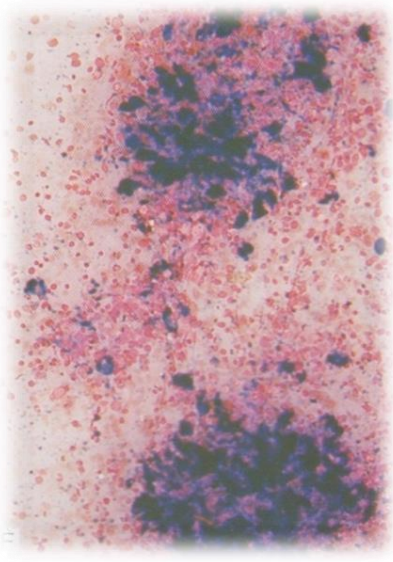


Fig.19[20]: Grade 6 iron stores



## BONE MARROW BIOPSY

| | Table 5: Indications for Bone marrow biopsy[18] |
|---|---|
| 1 | Diagnosis and/or staging of suspected Hodgkin lymphoma and non-Hodgkin lymphoma |
| 2 | Hairy cell leukemia |
| 3 | Chronic lymphocytic leukemia and other leukemic lymphoproliferative disorders |
| 4 | Diagnosis of suspected metastatic carcinoma |
| 5 | Diagnosis, staging, and follow-up of small cell tumors of childhood |
| 6 | Chronic myeloproliferative disorders (chronic myelogenous leukemia, polycythaemia rubra vera, essential thrombocythemia, idiopathic myelofibrosis, and mastocytosis) |
| 7 | Diagnosis of aplastic anemia |
| 8 | Investigation of an unexplained leukoerythroblastic blood smear |
| 9 | Investigation of a fever of unknown origin and/or granulomatous |
| 10 | Infection |
| 11 | Investigation of suspected hemophagocytic syndrome |
| 12 | Evaluation of any patient in whom an adequate bone marrow aspirate cannot be obtained |
| 13 | Suspected multiple myeloma or plasma cell dyscrasia |
| 14 | Suspected acute myeloid leukemia |
| 15 | Suspected myelodysplastic syndrome |
| 16 | Investigation of suspected storage disease |
| 17 | Suspected primary amyloidosis |

**BIOPSY SITES**[14]

The site chosen is determined by four key factors.:

1. the biopsy site should be easily accessible, involving a minimum of trauma and danger to the patient;

2. it should provide representative bone structure and turnover;

3. it should contain cortical and trabecular bone; and

4. repeat biopsies with minimal variability should be possible.

The ilium comes nearest to fulfilling these criteria, and the anterior and posterior iliac crest are the preferred sites from which the biopsies are taken

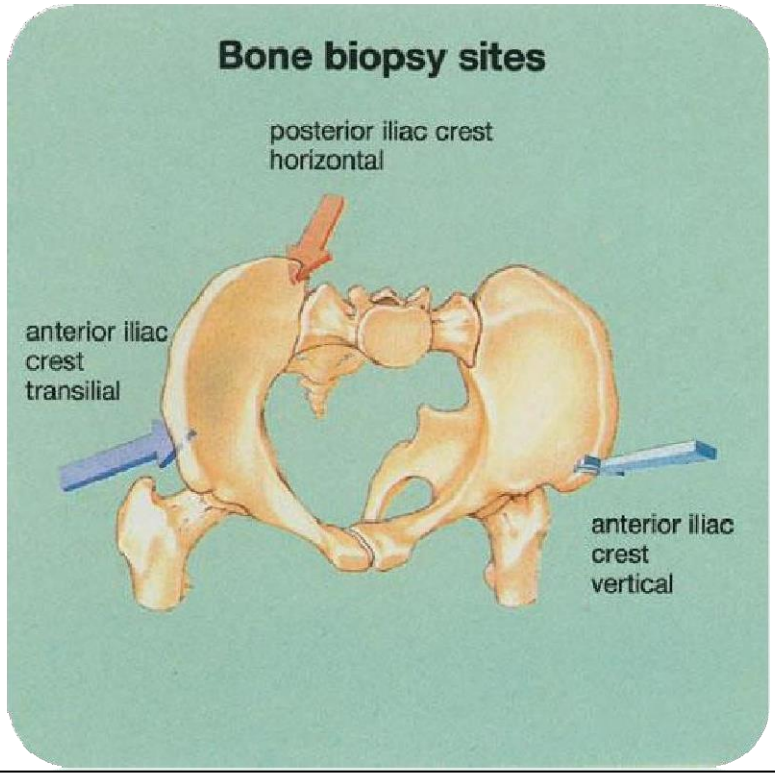



Fig.20[14] Sketch illustrating the three sites on the iliac crest from which bone biopsies are obtained

**BIOPSY INSTRUMENTS**[14]

There are two main kinds of instruments, namely the electric drill and the manual trephine, various types of which are commercially available.

The drill is used for vertical biopsies from the anterior iliac crest.**Fig.21**

The wide-bore manual trephine for horizontal transilial biopsies. **Fig 22**

Both provide relatively wide cores (4 mm and 8 mm respectively).

These are recommended when detailed histomorphometric measurements of cortical and trabecular bone are necessary, but at the cost of greater invasiveness and rate of complications. The anterior iliac crest biopsy cores, which are 4 mm in diameter and 20 mm long, have proven to be ideal sections for both qualitative and quantitative analysis. However, an 8-gauge manual trephine is used to collect the great majority of bone samples from the posterior iliac crest (3 mm width). This needle technique is especially appropriate for outpatients because it is less invasive, can be executed fairly easily, and has a range of lengths up to 70 mm.

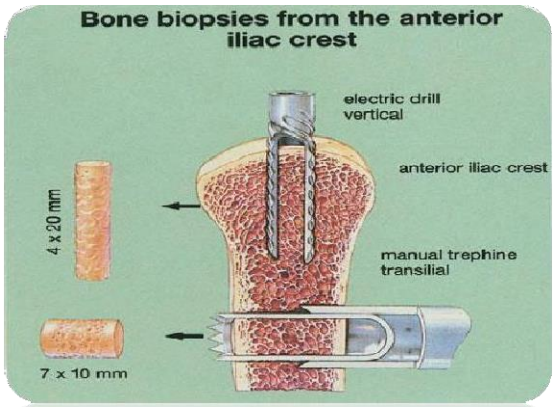



Fig.21[14]: Sketch of electric drill and manual transilial trephine in situ in the anterior ilium. Note progressive narrowing of the ilium with distance from the crest

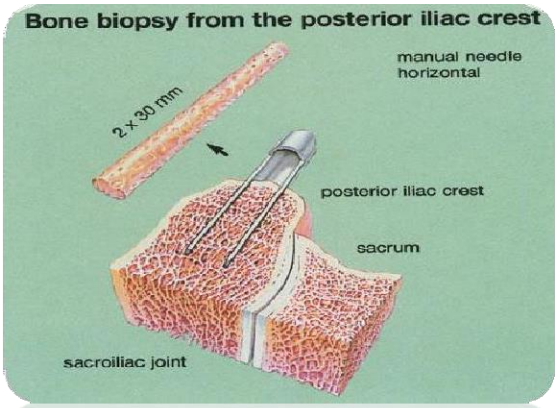



Fig.22[14]**:** Sketch of manual trephine in situ in the posterior ilium. Note width of iliac crest and distance of the needle from the sacroiliac

## DIAGNOSTIC BIOPSY EVALUATION [14]

Sections of biopsies demonstrates the range of normal quantitative relationships between cortical and trabecular bone, haematopoietic and adipose tissue. Part of the anterior and posterior cortex of the iliac crest is porous and of variable in thickness. Moreover, the thickness of the ossicles, as well as the structure of the trabecular network, may be variable even within a single section. It is important to consider the bone and marrow together as a single system, because alterations in the one are generally accompanied by changes in the other. It is also important to bear these considerations in mind, especially when histomorphometric measurements, comparative and follow-up studies are made and conclusions for therapy drawn from them. The main pitfalls in histological diagnosis are:

1. non-representative tangential biopsies, which are encountered more frequently when biopsies are taken from the narrow anterior iliac crest;

2. presence of misleading artefacts by crushing, distortion, and inclusion of other tissues (dermis, muscles); and

3. histological variations within the same biopsy, with subcortical hypoplasia or alternating fatty and hyperplastic areas in deeper parts of the biopsy, which in turn may affect the bone structure and remodeling.

## EVALUATION OF BONE MARROW EXTRACELLULAR STROMA, MARROW FIBROSIS, AND MESENCHYMAL CELLS

The extracellular matrix is demonstrable in routine preparations by reticulin (Gomori) silver stain, which stains most types of collagen including collagen III and collagen IV and by collagen IV immunostain, which stains the basal membrane collagen type. Bone marrow reticulum cells, also termed adventitial reticular cells, can be identified by their nerve growth factor receptor positivity (NGFR).

Trichrome stain (e.g.,Masson's) is commonly used to demonstrate the presence of "mature" collagen, which can occur in advanced stages of marrow fibrosis (i.e., osteomyelosclerosis). The grading of fibrosis, as a general rule, should be performed taking into account only areas of active hematopoiesis (fatty areas are excluded). In pathological bone marrow, areas of prominent stroma alterations, such as those with marked oedema or extensive fibrosclerosis, should also be included in the overall grading of the myelofibrosis.

**Table 6: Evaluation of fibrosis**

| | |
|---|---|
| 1+ | Focal fine fibres with only rare coarse fibres |
| 2+ | A diffuse fine fibre network with an increase in scattered coarse fibres |
| 3+ | A diffuse coarse fibre network with no collagenization (negative trichrome stain) |
| 4+ | A diffuse fibre network with collagenization (positive trichrome stain) |

## MATERIALS AND METHODS:

• Source of data

All patients requiring Bone marrow aspiration and biopsy with diagnosis of pancytopenia referred to the hematology laboratory of Pathology department, BLDE (DEEMED TO BE UNIVERSITY) Shri B. M. Patil Medical College, Hospital and Research Centre, Vijayapura was included in the study.

Study period: 1st January 2021 to 31st July 2022

7.2 Methods of collection of data.

Complete clinical data was recorded, including physical examination, complete Blood counts using sysmex XN 1000, together with other relevant investigations within the proforma. Patients requiring bone marrow examination was selected for the study.

Bone marrow Aspiration, imprint and biopsy was performed as per the following procedure.

PROCEDURE:[18]

Explanation of the procedure and written consent was obtained prior to the procedure.

1. Posterior superior iliac spine is the most satisfactory and safe site for both aspiration and biopsy.

2. Explanation about the procedure to patients was done and made to lie either in the left or right lateral position with the knees drawn up .

3. The skin in the area of the procedure (posterior iliac spine) was cleaned with 70% ethanol and betadine. The skin, subcutaneous tissue, and periosteum was infiltrated with 2ml of 2% Xylocaine(lignocaine) as a local anesthetic. With a boring movement, the Jamshidi trephine biopsy needle was passed perpendicularly into the cavity of the bone.

The stilette was then removed, and 0.2-0.3ml of marrow contents was sucked with the help of a 10ml syringe and delivered into a bottle containing EDTA to make films later and stained with Leishman's stain. The same biopsy needle was inserted into the bone using to-and-fro rotation to obtain a core of tissue. Benzoin seal was applied on the puncture site.

4. The specimen was fixed in Bouin's solution and decalcified with 9.5% of nitric acid in 1% EDTA for 48 hrs.

Before fixation of the biopsy minimum of five touch imprint smears were prepared by using the procedure of gentle touch and rolling of the biopsy core on the slide. After routine processing and paraffin embedding, hematoxylin and eosin staining was studied. Special stains like Perl's Prussian Blue stain, Reticulin, and Myeloperoxidase (MPO) were done whenever necessary.

Sample size:

With the anticipated Proportion of Pancytopenia among Hematological disorders patients 26.92% (ref) in the population, the study would need a sample size of a minimum of 53 patients with a 95% level of confidence and 10% absolute precision.

Formula used

• $n=z2\ p*q$

$d2$

Where Z= Z statistic at α level of significance

d2= Absolute error

P= Proportion rate

q= 100-p

• Statistical Analysis

• The data obtained was entered in a Microsoft Excel sheet, and statistical analysis was done by statistical package for the social sciences (Version 20).

• Results were presented as Mean (Median) ±SD, counts and percentages, and diagrams.

• Comparison of different diseases were made using the Chi-square test.

$P<0.05$ will be contemplated as statistically significant.

Inclusion criteria:

• All the cases with pancytopenia (hemoglobin <9g/dl, total leucocyte count < 4x10 9 /L, and platelet count <140x10 9 /L) requiring Bone marrow aspiration, imprints, and biopsy examination were included in the study.

Exclusion criteria:

• Biopsy <0.5 cm were excluded from the study.

## OBSERVATIONS AND RESULTS:

A total of 64 patients with pancytopenia were evaluated. Sex distribution among various age groups, peripheral smear findings, bone marrow aspiration, imprint smear and biopsy were analyzed.

AGE GROUP

Table7: Incidence of pancytopenia in different age groups

| | | Frequency | Percent |
|---|---|---|---|
| | 01-12yrs | 9 | 14.1 |
| | 13-22yrs | 14 | 21.9 |
| | 23-32yrs | 15 | 23.4 |
| | 33-42yrs | 7 | 10.9 |
| | 43-52yrs | 8 | 12.5 |
| | 53-62yrs | 5 | 7.8 |
| | 62-72yrs | 4 | 6.3 |
| | 75+yrs | 2 | 3.1 |
| | Total | 64 | 100.0 |

| AGE STATISTICS | | |
|---|---|---|
| Mean | | 32.81 |
| Median | | 29.50 |
| Std. Deviation | | 19.346 |
| Percentiles | 25 | 18.50 |
| | 50 | 29.50 |
| | 75 | 45.00 |

The commonest age group affected was 23-32(23.4%) years and least affected age group was 75-83 (3.1%).

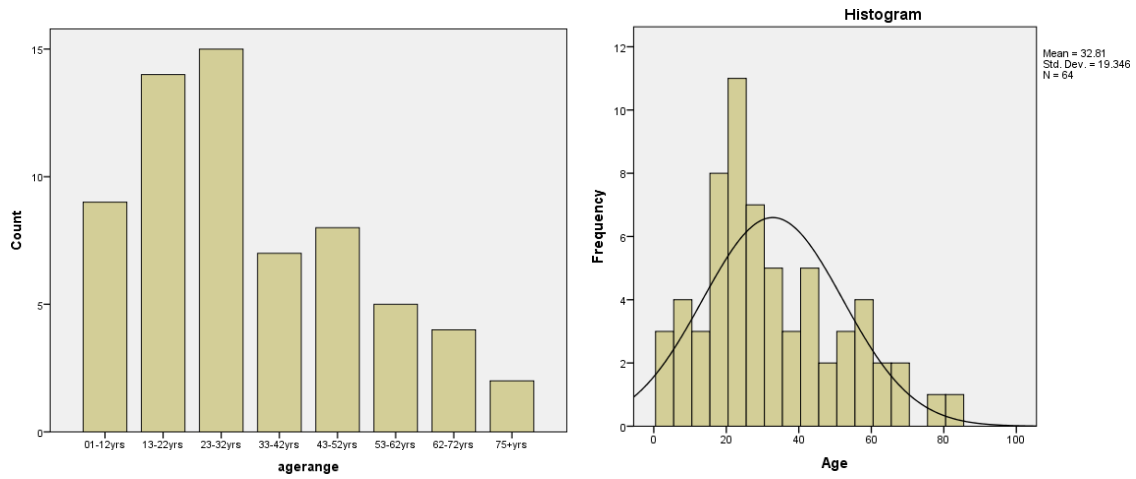



Fig 23: Incidence of pancytopenia in different age groups

SEX DISTRIBUTION

Table 8: Incidence of pancytopenia in different sex groups

| | | Frequency | % |
|---|---|---|---|
| | Female | 27 | 42.2 |
| | Male | 37 | 57.8 |
| | Total | 64 | 100.0 |

In our study male predominance was noted with males accounting to 57.8% and females 42.2%.

Fig 24: Incidence of pancytopenia in different sex group

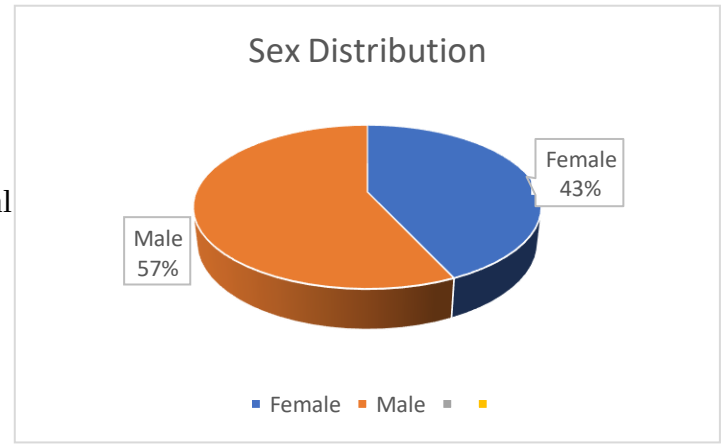



Table 9: Peripheral

| | Frequency | % |
|---|---|---|
| Dimorphic anemia | 20 | 31.3 |
| Macrocytic anemia | 17 | 26.6 |
| MCHC anemia | 8 | 12.5 |
| NCNC anemia | 19 | 29.7 |
| Total | 64 | 100.0 |

In present study dimorphic anemia was seen in majority of cases (31.3%), followed by normocytic normochromic anemia (29.7%), macrocytic anemia(26.6%) and few cases showed microcytic hypochromic anemia(12.50%).

Fig.25: Peripheral Blood smear in pancytopenia

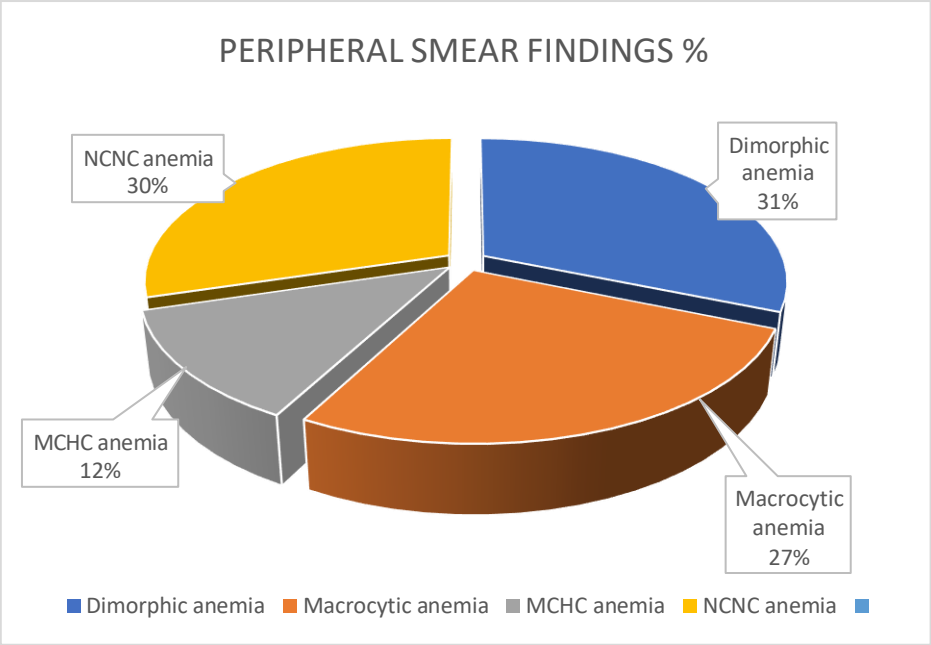



Table 10: Bone marrow cellularity

| Cellularity | Number of Cases | % |
|---|---|---|
| HYPERCELLULAR | 54 | 84.38% |
| HYPOCELLULAR | 7 | 10.94% |
| NORMOCELLULAR | 3 | 4.69% |
| Grand Total | 64 | 100.00% |

In present study, 84.38 % cases had hypercellular marrow. Megaloblastic anemia was common cause for hypercellularity. 10.94% and 4.69% of cases were hypocellular and normocellular respectively.

Fig. 26: Bone marrow cellularity

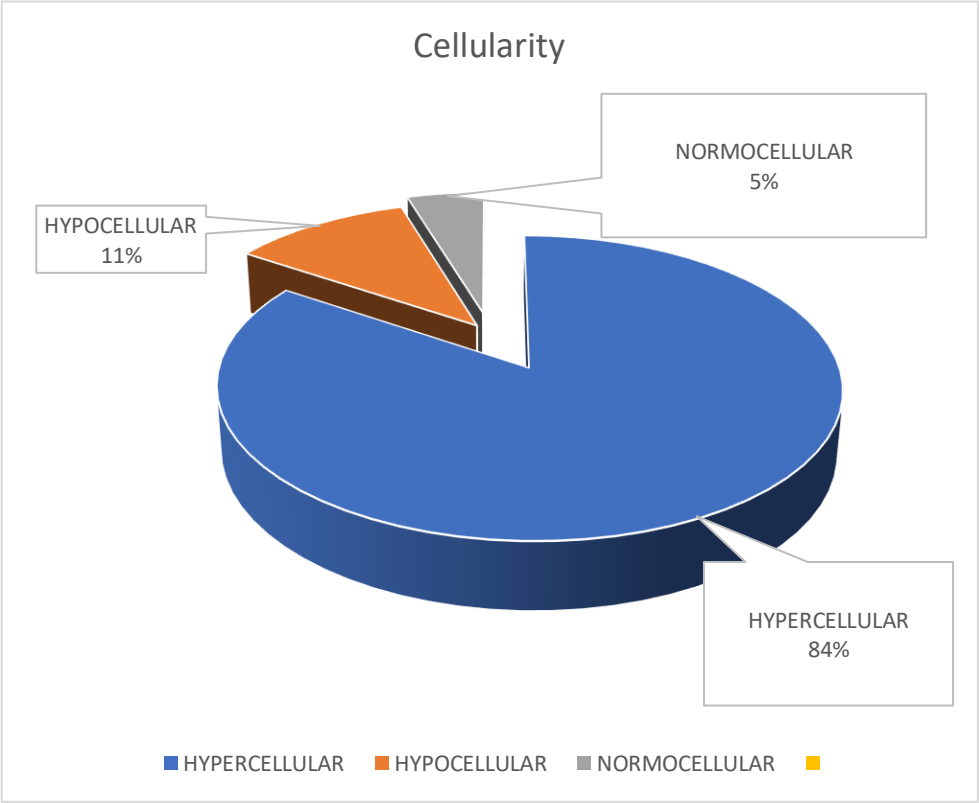
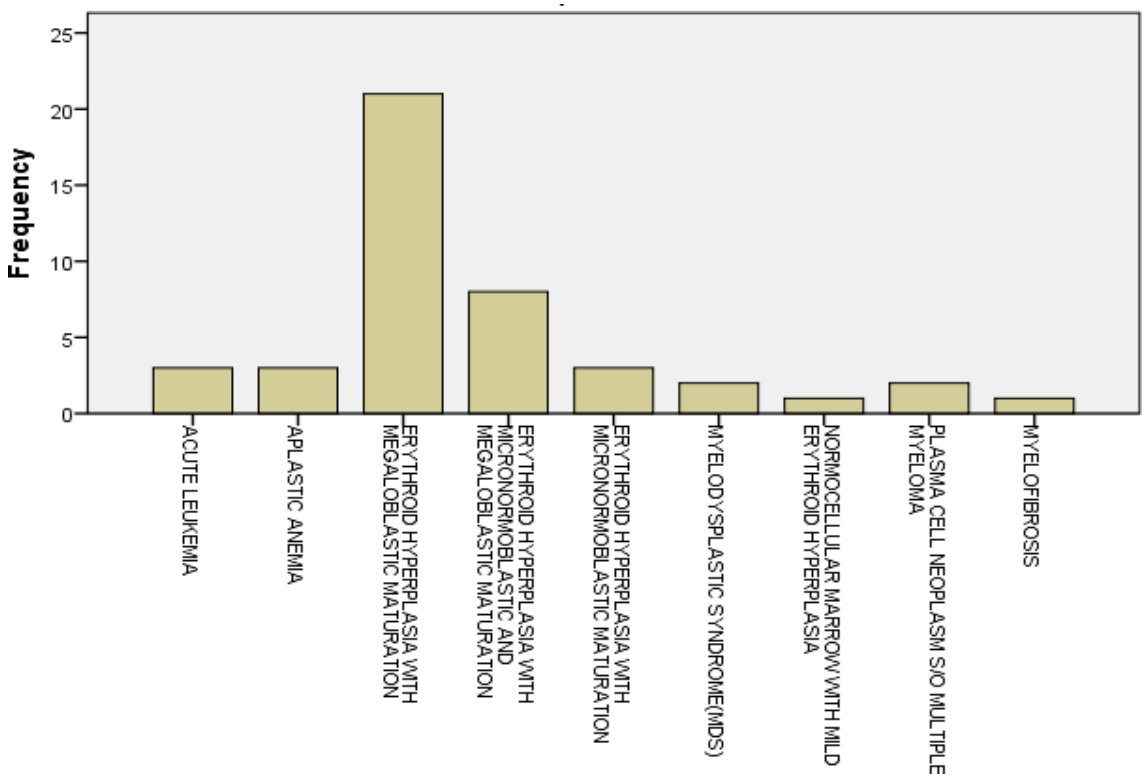



Table 11: Distribution of various causes of pancytopenia

| Distribution of various causes of Pancytopenia | Frequency | % |
|---|---|---|
| ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | 21 | 32.8 |
| ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | 8 | 12.5 |
| ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | 6 | 9.4 |
| ACUTE LEUKEMIA | 3 | 4.7 |
| APLASTIC ANEMIA | 3 | 4.7 |
| PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | 2 | 3.1 |
| MYELODYSPLASTIC SYNDROME(MDS) | 2 | 3.1 |
| MYELOFIBROSIS | 1 | 1.6 |
| FILARIASIS | 1 | 1.6 |
| Total | 64 | 100.0 |

Fig.27: Distribution of various causes of pancytopenia




In the present study, megaloblastic anemia was commonest cause with 47.7% followed by erythroid hyperplasia with both micronormoblastic and megaloblastic maturation 18.2%. Least common cause were myelofibrosis, multiple myeloma, MDS and Filariasis.

**Table12: Pancytopenia Etiology Sex distribution**

| | | ACUTE LEUKEMIA | MEGALOBLASTIC MATURATION | MICRONORMOBLASTIC AND MEGALOBLASTIC | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC | MYELODYSPLASTIC SYNDROME(MDS) | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | MYELOFIBROSIS | HYPOCELLULAR MARROW | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC | Total |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Sex | Female | 2 | 10 | 6 | 3 | 0 | 0 | 0 | 2 | 2 | 27 |
| | Male | 2 | 20 | 2 | 3 | 2 | 3 | 1 | 3 | 0 | 36 |
| Total | | 4 | 30 | 8 | 6 | 2 | 3 | 1 | 5 | 2 | 63 |

**Table 13: Pancytopenia Etiology Age Distribution**

| Age range | ACUTE LEUKEMIA | MEGALOBLASTIC MATURATION | MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | MICRONORMOBLASTIC MATURATION | MYELODYSPLASTIC SYNDROME(MDS) | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | MYELOFIBROSIS | HYPOCELLULAR MARROW | NORMOBLASTIC MATURATION | Total |
|---|---|---|---|---|---|---|---|---|---|---|
| 01-12yrs | 2 | 1 | 0 | 1 | 0 | 0 | 0 | 3 | 0 | 7 |
| 13-22yrs | 1 | 10 | 2 | 0 | 0 | 0 | 0 | 0 | 1 | 14 |
| 23-32yrs | 0 | 9 | 3 | 1 | 0 | 0 | 0 | 1 | 0 | 14 |
| 33-42yrs | 0 | 2 | 1 | 3 | 1 | 0 | 0 | 0 | 0 | 7 |
| 43-52yrs | 0 | 5 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 8 |
| 53-62yrs | 1 | 3 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 5 |
| 62-72yrs | 0 | 0 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 5 |
| 75+yrs | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 2 |
| Total | 4 | 30 | 8 | 6 | 2 | 3 | 3 | 5 | 2 | 63 |

MEGALOBLASTIC ANEMIA ASSOCIATED WITH PANCYTOPENIA

Out of 64 cases, 31 were megaloblastic anemia, the commonest age group affected was 12-23 yrs. Males (62.33%) affected more than females (37.66%). Peripheral study had dimorphic picture in most cases (72.72%). Bone marrow study had hypercellularity in 89.16% cases of megaloblastic anemia (Fig.23).

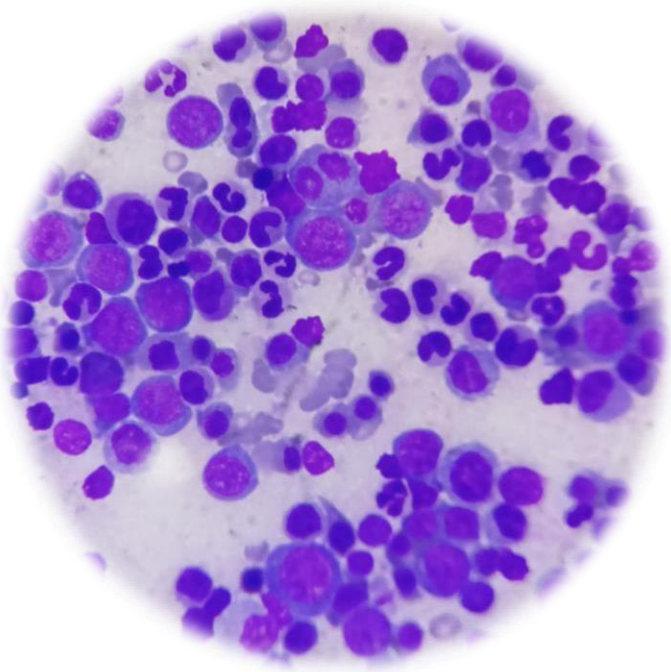


Fig.28: Microphotograph demonstrating megaloblastic anemia.

IRON DEFICIENCY ANEMIA ASSOCIATED WITH PANCYTOPENIA

Six cases of iron deficiency anemia was noted with commonest age group affecting 15-25yrs and female show preponderance.

Bone marrow study of most cases were hypercellular with altered M: E ratio, increased erythropoiesis showing micronormaoblasts. Myelopoiesis was normal and there was slight increase in megakaryocytes.

Bone marrow iron stores were within 0-2 grade that is complete absent to reduced iron stores.

LEUKEMIA ASSOCIATED WITH PANCYTOPENIA:

In the present study, 4 patients had hypercellular marrow due to leukemia, out of which 3 cases were CLL and one case was AML.

In all cases of CLL, peripheral smear showed features of microcytic hypochromic anemia with neutropenia and thrombocytopenia. 90% were mature lymphocytes with presence of many smudge cells.

BM was hypercellular with presence of 80% mature lymphocytes with smudge cells. Erythroid, myeloid and megakaryocytic series were reduced.

In case of AML peripheral smear showed microcytic hypochromic anemia with neutropenia and thrombocytopenia. Myeloblasts constituted 20% of differential count with few showing auer rods. BM was hypercellular. Erythroid and megakaryocytic series were reduced. Majority of cells were myeloblasts constituting 40% of cells in marrow.

MPO stain on bone marrow aspirate smears showed 30% blast positivity.

APLASTIC ANEMIA ASSOCIATED WITH PANCYTOPENIA:

In present study 3 cases were diagnosed as aplastic anemia.

Peripheral smear was normocytic normochromic anemia with neutropenia and thrombocytopenia.

Bone marrow aspirate was hypocellular with increased fat cells.

Imprint smears helped in accessing the cellularity when aspiration smears were inadequate for opinion.

Bone marrow biopsy showed increased adipose tissues in intertrabecular spaces with relative increase in plasma cells and lymphocytes. All three lineages were reduced in number (Fig.29).

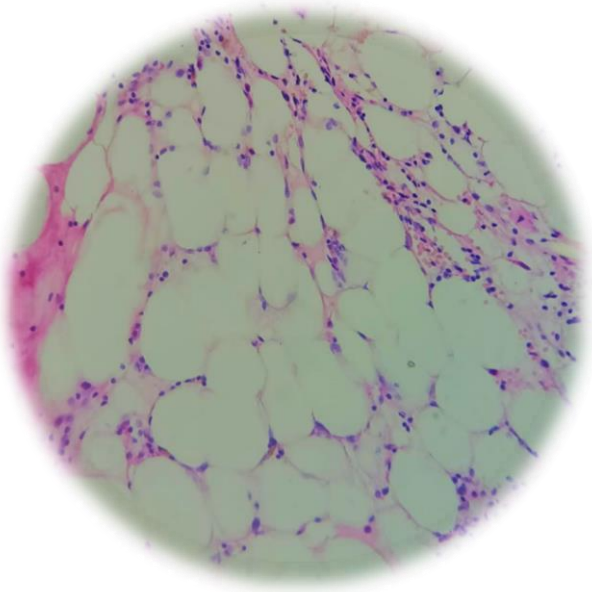


Fig.29: Microphotograph showing increase adipocytes with decrease in all 3 lineages.

MULTIPLE MYELOMA PRESENTING AS PANCYTOPENIA:

Three cases were diagnosed as Multiple myeloma.

Peripheral smear findings on multiple myeloma was predominantly normocytic normochromic anemia with rouleaux formations.

BM examination was satisfactory and hypercellular. Erythropoiesis, leucopoiesis and megakaryopoiesis were suppressed. There was abnormal proliferation of plasma cells >40%, showing binucleate and trinucleate forms.

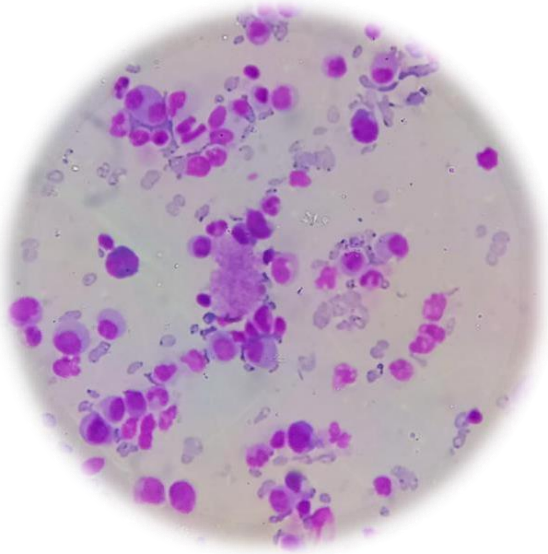


Fig.30: Microphotograph showing proliferation of plasma cells

MYELOFIBROSIS ASSOCIATED WITH PANCYTOPENIA:

One case was diagnosed as myelofibrosis.

Peripheral smear showed microcytic hypochromic anemia.

Bone marrow aspirate was dry tap.

Imprint and clot preparations showed presence of microfilaria.

Bone marrow biopsy showed increased fibrosis, no presence of parasite was noted on biopsy.

Reticulin stain: Increased fibrosis.

Fig.31 Bone marrow clot preparation

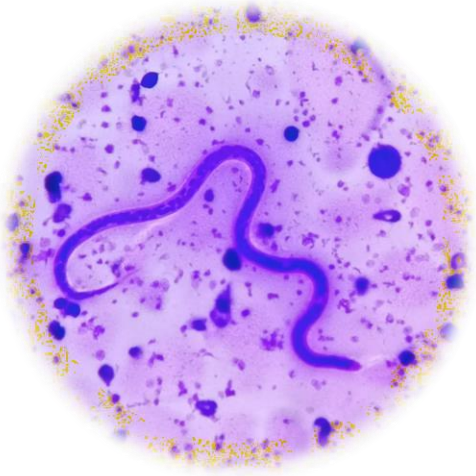
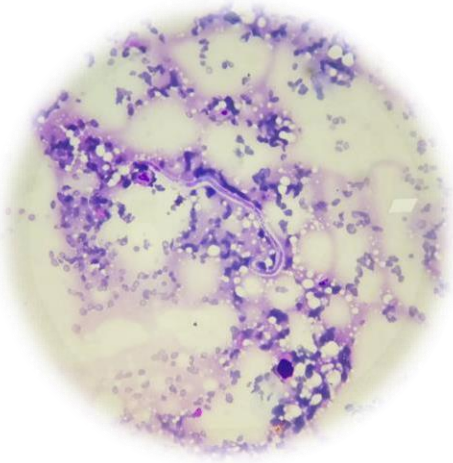


Fig.32 Bone marrow imprint showing microfilaria



MDS ASSOCIATED WITH PANCYTOPENIA:

2 cases were diagnosed as Myelodysplastic syndrome. Peripheral smear of one case showed macrocytic anemia with ?10% blast/Atypical lymphocyte. Bone marrow aspiration was normocellular with erythroid hyperplasia and megaloblastic maturation. 10% dyserythropoietic cells were noted showing karyorrhexis, cytoplasmic irregularity with basophilic granules.

Bone marrow biopsy showed hypercelluar marrow with erythroid hyperplasia and megaloblastic maturation. Abnormal localization of myeloid precursors

were noted in the intertrabecular spaces of marrow. Few hypolobated and micromegakaryocytes were noted. So it was reported as features suggestive of MDS-Single lineage dysplasia and cytogenetic study was suggested.

Peripheral smear finding of other case was dimorphic anemia. Bone marrow aspirate showed hypercellular marrow with increased erythropoiesis. Dyserythopoiesis was noted in more than >10% of erythroid series. Myeloid series showed hypogranular myelocytes, metamyelocytes, band forms and neutrophils with presence of ring neutrophils. Few hypolobated and micromegakaryocytes were noted. So it was reported as features suggestive of MDS-Multilineage dysplasia and cytogenetic study was suggested.

Table 14: Descriptive Statistics BMA/ BMI/ BMB

| | Mean | Std. Deviation | N |
|---|---|---|---|
| BMA Impression | 4.00 | 2.365 | 45 |
| BMI Impression | 3.69 | 1.844 | 45 |
| BMB Impression | 3.58 | 1.588 | 45 |

| Table 15: DIAGNOSTIC ACCURACY OF BMA, BMI & BMB | | | |
|---|---|---|---|
| IMPRESSION | BMA | BMI | BMB |
| ACUTE LEUKEMIA | 6.3 | 6.3 | 6.3 |
| APLASTIC ANEMIA | 40.6 | 4.7 | 4.7 |
| ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | 12.5 | 45.3 | 48.4 |
| ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | 7.8 | 9.4 | 12.5 |
| ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | 3.1 | 7.8 | 7.8 |
| MYELODYSPLASTIC SYNDROME(MDS) | 3.1 | 1.6 | 3.1 |
| NORMOCELLULAR MARROW WITH MILD ERYTHROID HYPERPLASIA | 7.8 | 3.1 | 10.9 |
| PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | 3.1 | 3.1 | 4.7 |
| MYELOFIBROSIS | 0 | 4.7 | 1.6 |
| Total | 84.4 | 85.9 | 100.0 |

Table 16: Bone marrow aspiration imprint and biopsy correlation

| Control Variables | | | BMA Impression | BMI Impression | BMB Impression |
|---|---|---|---|---|---|
| | BMB Impression | Correlation | .769 | .994 | 1.000 |
| | | Significance (2-tailed) | .000 | .000 | |
| | | df | 43 | 43 | 0 |
| | BMA Impression | Correlation | 1.000 | .328 | |
| | | Significance (2-tailed) | | .030 | |
| | | df | 0 | 42 | |
| | BMI Impression | Correlation | .328 | 1.000 | |
| | | Significance (2-tailed) | .030 | | |
| | | df | 42 | 0 | |

The present study observed that although the diagnostic accuracy of BMB was highest (100%) but diagnostic accuracy of BMI was also considerably high (85.9%) in comparison to BMA (84.4%) in diagnosing various pancytopenia cases.

Current study showed a positive correlation with a p value of 0.03.

# DISCUSSION

Pancytopenia is a common haematological entity encountered in clinical practise. A thorough clinical, haematological, and bone marrow examination of patients usually aids in the identification of the underlying cause. However, due to a variety of etiological variables, pancytopenia remains a difficulty. When done appropriately, bone marrow aspiration is an indispensable tool in the research of haematological diseases. It is simple, safe, and repeatable. The larger advantage of trephine biopsy is that it can provide information on the structure of quite big pieces of marrow. Simultaneously, morphological properties of individual cells can be detected by making an impression from the collected material. As a result, the current study was designed to assess the accuracy of bone marrow aspiration, imprint and biopsy in the diagnosis of haematological illnesses characterised by pancytopenia.

In order to investigate the numerous causes of pancytopenia, we studied 64 pancytopenia cases. Its age and sex distribution, with peripheral smear examination, associated marrow manifestations and comparative analysis of bone marrow aspiration, imprint smear and biopsy was evaluated. The obtained statistical data were compared to previously published literature.

**TABLE 17: AGE DISTRIBUTION IN COMPARISON WITH OTHER STUDIES**

| Sl no. | Authors | Common age group affected | Total No. Of cases |
|---|---|---|---|
| 1 | Bhuyan A et *al*[21](2022) | 18-28 | 42 |
| 2 | Halder B *et al*[22](2022) | 20-30 | 62 |
| 3 | Singh B et al[23] (2021) | 11-20 | 50 |
| 4 | Pasam R *et al*[24](2016) | 21-30 | 28 |
| 5 | Metikurke SH *et al*[11](2013) | 21-30 | 58 |
| 6 | Present Study | 23-32 | 64 |

The distribution of ages and genders was comparable to other pancytopenia investigations. 1st to 3rd decade of life were often impacted in the studys conducted by Bhuyan A e*t al*[21], Halder B *et al*[22], Singh B et al[23], Pasam R et al, Metikurke SH *et al*[11]

Age group 23–32 years was most typically impacted in the current study (table 7).

Males are affected more than females in all studies. In the current survey, men outnumber women by a ratio of 1.32:1.

**TABLE 18: SEX DISTRIBUTION IN COMPARISON WITH OTHER STUDIES**

| Sl No: | Authors | M:E Ratio |
|---|---|---|
| 1 | Bhuyan A e*t al*[21] (2022) | 1.33:1 |
| 2 | Taori G at el[25](2019) | 1.01:1 |
| 3 | Reddy GPK et al[26](2016) | 1.2:1 |
| 4 | Pathak R et al[12](2012) | 1:1.04 |
| 5 | Gayatri BN et al[6] (2011) | 1.2:1 |
| 6 | Present Study | 1.35:1 |

In all the comparable studies males were found to be affected more than females. Our study also showed male predominance with male: female ratio of 1.35 : 1.

**Table 19: DISTRIBUTION OF PANCYTOPENIA ETIOLOGY COMPARISON WITH OTHER STUDIES**

| Study | No. of patients | Commonest cause | Other common causes | Rare cause |
|---|---|---|---|---|
| Gayatri BN[6] *et al* [6] | 104 | MA (74.04%) | AA (18.26%), Leukemia (3.85%) | Malaria, MM, Storage disorder. |
| Khunger JM *et al* [9] | 200 | MA (72%) | Aplastic anemia (14%), Leukemia (5%) | Malaria, KZ, NHL, MF, MM, dTB |
| Javalgi AP *et al* [26] | 106 | MA (72.6%) | IDA (12.4%), Malaria (3.7%) | Leukemia, SLE, AA, MM, MF, MDS, HS |
| Santra G *et al* [27] | 111 | AA (22.72%) | Hypersplenism, CLD, KZ, Mixed deficiency anemia (6.31%). | Malaria, dTB, NHL, CLL, Leukemia, MDS |
| Reddy GPK *et al*[26] | 42 | MA (38.1%) | Aplastic anemia (26.2%) | AML, Malaria, Malignancy, ALD, MM, Tuberculosis |

| | | | | |
|---|---|---|---|---|
| Tejaswini V *et al* [28] | 75 | MA (68%) | Aplastic Anemia (13.3%) | Leukemia, Myelofibrosis, ITP, MM |
| Present study | 62 | MA (47.7%) | Mixed deficiency anemia (12.5%), IDA (9.4%), | AA, AL, MF, Plasma cell neoplasia, Filariasis |

According to Khunger et al.[3] and Tilak et al.[9], megaloblastic anemia was the most common cause of pancytopenia in their study, accounting for 50 percent of cases.

In our study, erythroid hyperplasia with megaloblastic maturation (47.7%) was the common finding followed by erythroid hyperplasia with micronormoblastic and megaloblastic maturation (12.5%) which was the second most common contributor to pancytopenia, in stark contrast to other authors studies. This appears to be due to Indian subjects having a higher prevalence of nutritional anaemia. [5, 9]

In their study of 100 patients with panytopenia, Osama Ishtiaq et al.[7] found five cases of iron deficiency anaemia (5%), which was equivalent to our study in which we also found 6 cases of iron deficiency anaemia presenting as pancytopenia. Pancytopenia may be related to iron deficiency anaemia. Even though reactive thrombocytosis is linked to iron deficiency, the severity of the condition causes platelet counts to normalise and, on rare occasions, even decline. [22] Although the precise process is unknown, it may be connected to the altered activity of iron-dependent enzymes during thrombopoiesis and leucopoiesis.[23].

In the investigations conducted by Tilak et al.[9] and Khunger et al.[3], aplastic anaemia was the second most prevalent cause, but in the current study, there were only three cases, indicating that the prevalence of the condition varies.

In contrast to Khunger JM et al[3] who also reported 1% cases of multiple myeloma in their study, we had 2 cases of multiple myeloma, with an incidence of 6.45%.

Similar to the current study, which also had one case of myelofibrosis with pancytopenia, Khunger et al.[3]'s study found 1% cases of myelofibrosis.

AML, hairy cell leukaemia, multiple myeloma, myelofibrosis, hemophagocytic syndrome, tuberculosis, multiple myeloma and drug-induced pancytopenia were uncommon causes of pancytopenia in the studies by Khunger JM et al[3] and Tilak et al[9]. Myelofibrosis, MDS, and multiple myeloma were uncommon causes of pancytopenia in the present study.

One case was diagnosed as myelofibrosis on biopsy where aspiration was dry tap. One of the imprint showed incidental finding of microfilaria and bone marrow biopsy gave a diagnosis of myelofibrosis. In a case study done by Khaliqur Rahman et al[18], discussed about association of microfilariae with MPN or myelofibrosis which corelated to the clinal scenarios which we observed in our case. Further investigations and follow up was limited.

**Table 20: DIAGNOSTIC ACCURACY OF BONE MARROW ASPIRATION, IMPRINT AND BIOPSY COMPARED TO OTHER STUDIES**

| | Present Study | Chandra S *et al*[29] | Pant S *et al*[30] | Taori G *et al*[25] |
|---|---|---|---|---|
| Total number of cases | 63 | 63 | 565 | 111 |
| BMA | 54(84.4%) | 53(84%) | 438(75%) | 88(79%) |
| BMI | 55(85.9%) | 60(95%) | 473(83%) | 97(87%) |
| BMB | 63(100%) | 63(100%) | 561(99%) | 111(100%) |

Pancytopenia has either hypercellular or hypocellular bone marrow morphology, and there are only a few research that examine the many etiological variables of pancytopenia with either cellular or hypocellular marrow.

# SUMMARY


- A present study "A Comparative Study of Bone Marrow Aspiration, Imprint and Biopsy in Pancytopenia" was undertaken at Shri B.M Patil Medical college, Vijayapura.
- Total sixty-four patients of pancytopenia requiring bone marrow study of all age groups were evaluated.
- A combined evaluation of primary hematological investigations combined with bone marrow aspiration, imprint and biopsy were done.
- Commonest age group affected was 23-32 yrs followed by 13-22 yrs.
- Males predominance was observed in this study with a Male : Female ratio of 1.35:1.
- Peripheral smear picture was dominated by dimorphic anemia.
- Erythroid hyperplasia with megaloblastic maturation (30 cases) was the most commonest finding, followed by erythroid hyperplasia with micronormoblastic and megaloblastic maturation (8 cases) and erythroid hyperplasia with micronormoblastic maturation (6 cases) on bone marrow examination.
- Other causes of pancytopenia were aplastic anemia, acute leukemia, multiple myeloma and myelodysplastic syndrome.
- Rare causes of pancytopenia were myelofibrosis and filariasis.
- Bone marrow examination showed predominantly hypercellular marrow (84.38%) followed by hypocellular (7%) and normocellular marrow (4.69%).
- All the three procedures of bone marrow aspiration, touch imprint and trephine biopsy were found to be complementary to each other and superiority of one method over the other depended on the specific disease process.

# CONCLUSION

Pancytopenia is a common haematological problem encountered in clinical practice and should be suspected on clinical grounds when a patient presents with unexplained anaemia, prolonged fever and tendency to bleed.

The physical findings and peripheral blood picture provides valuable information in the work of cytopenic patients.

Evaluation of peripheral blood film reveals the most probable cause of anaemia, presence of nucleated RBC's and/or immature myeloid cells may suggest marrow infiltration or primary haematologic disorder.

Bone marrow aspiration is an important diagnostic tool in haematology which helps to evaluate various cases of cytopenia.

Bone marrow examination is accurate, reproducible, rapidly available information at an economical cost and with minimal discomfort to the patient. Bone marrow aspiration is sufficient to make a diagnosis in cases of nutritional anaemias and initial diagnosis of leukemia.

Megaloblastic anaemia was the commonest cause which indicates the high prevalence of nutritional anaemia in our region.

The other common causes were combined nutritional anemia, aplastic anemia and leukemia.

However, uncommon and rare causes such as multiple myeloma, storage disease should be kept in mind while planning investigation for complete work up of cytopenic patients.

Tuberculosis being highly prevalent and endemic in India, it is essential to be aware of its manifestation as pancytopenia.

Present study concludes that numerous causes of pancytopenia which include both non neoplastic and neoplastic conditions should be evaluated by clinical findings and peripheral smear with Bone marrow examination which includes BMA, BMI and BMB is an essential pre requisite for its diagnosis. Though the advantage of each procedure differs, both the procedures are complimentary to each other and should be performed simultaneously along with an imprint smear for a complete bone marrow workup and evaluation. BMI are unquestionably important for investigation of bone marrow pathologies. It provides better information regarding marrow cellularity in comparison to BMA in cases of dry tap.

Owing to less mutilation, cellular cytological details are better appreciated in BMI. Even it provides some important diagnostic clues (e.g. marked adipocytosis). All these are ultimately reflected in better diagnostic accuracy of BMI as compared to BMA. It also avoids the unnecessary delay caused by decalcification and processing of BMB sections in routine histopathology laboratories.

# REFERENCE


1. Vargas-Carretero CJ, Fernandez-Vargas OE, Ron-Magaña AL, Padilla-Ortega JA, Ron-Guerrero CS, Barrera-Chairez E. Etiology and clinico-hematological profile of pancytopenia: experience of a Mexican Tertiary Care Center and review of the literature. Hematology. 2019 Dec;24(1):399-404. [PubMed].
2. Das Makheja K, Kumar Maheshwari B, Arain S, Kumar S, Kumari S, Vikash The common causes leading to pancytopenia in patients presenting to tertiary care hospital. Pak J Med Sci. 2013 Sep;29(5):1108-11. [PMC free article] [PubMed]
3. Santra G, Das BK. A cross-sectional study of the clinical profile and aetiological spectrum of pancytopenia in a tertiary care centre. Singapore Med J. 2010 Oct;51(10):806-12. PMID: 21103817.
4. Keisu M, Ost A. Diagnoses in patients with severe pancytopenia suspected of having aplastic anemia. Eur J Haematol. 1990 Jul;45(1):11-4. doi: 10.1111/j.1600-0609.1990.tb00407.x. PMID: 2379559.
5. Desalphine M, Bagga PK, Gupta PK, Kataria AS. To Evaluate the Role of Bone Marrow Aspiration and Bone Marrow Biopsy in Pancytopenia. Journal of clinical and diagnostic research : JCDR. 2014; 8(11).
6. Gayathri BN, Rao KS. Pancytopenia: A clinico hematological study. Journal of Laboratory Physicians. 2011; 3(1): 15-20.
7. Asam R, Garlapati, Chaganti P, Panchakarla G. — A Clinico-Hematological Study of Pancytopenia Ⅱ. IOSR Journalof Dental and Medical Sciences. 2016;15(5):71–6.
8. Gurkan UA, Akkus O. The Mechanical Environment of Bone Marrow: A Review. Annals of

Biomedical Engineering. 2008; 36(12): 1978-1991.

9. Khunger JM, Arulselvi S, Sharma U, Ranga S, Talib VH. Pancytopenia-A Clinico-haematological study of 200 cases. Indian J Pathol Microbiol. 2002; 45(3): 375-9.
10. Guinan EC, Shimamura A. Acquired and inherited aplastic anemia in: Greer JP, Lukens JN, Foerster J, Rodgers GM, Paraskavas F, Glader B(edt), Wintrobe's clinical hematology,11th ed.Lippincott Williams and Wilkins .2008:p1397-1402.
11. Metikurke SH, K. Rashmi, Rishi Bhavika R. Correlation of bone marrow aspirate, biopsies and touch imprint findings in pancytopenia. Journal of Hematology. 2013;
12. Pathak R, Jha a, Sayami G. Evaluation of bone marrow in patients with pancytopenia. J Pathol Nepal. 2012;22:265-75.
13. Ishtiaq O, Baqai HZ, Anwer F, Hussai N. Patterns of pancytopenia patients in ageneral medical ward and a proposed diagnostic approach. www.ayubmed.edu.pk/JAMC/PAST/16-1/osama.htm-206K-6/24/2007.
14. ISLAM ANWARUL. CHAPTER 2 BONE MARROW ASPIRATION. In: Manual of Bone Marrow Examination. S.l.: CAMBRIDGE SCHOLARS PUBLIS; 2022.
15. Boes KM, Durham AC. Bone Marrow, blood cells, and the lymphoid/lymphatic system. Pathologic Basis of Disease. 2017;
16. Kumar V, Abbas AK, Aster JC. Diseases of White Blood cells, Lymphnode, Spleen, and Thymus In: Robbins and Cotran Pathologic Basis of Disease. 9th ed. Philadelphia: Elsevier Saunders; 2015.p.579-81.
17. Wickramasinghe SN, Erber WN. Normal blood cells. Wickramasinghe SN, Porwit A, Erber WN. Normal bone marrow cells: development and cytology In: Porwit A, McCullough J, Erber W, editors. Blood and bone marrow pathology.2nd ed. Churchill

Livingstone/Elsevier.China;2011.

18. Bain BJ. Normal Bone Marrow. In: Bone Marrow pathology. Oxford: Blackwell Science; 2001.
19. Pujara K, Bhalara R, Dhruva G. A study of bone marrow iron storage in hematological disorder. Int J Heal Allied Sci 2014;3:221- 4. Available from: http://www.ijhas.in/text.asp?2014/3/4/221/143052
20. Singh T. Normal hematopoiesis and evaluation of bone marrow and peripheral blood film.In:Atlas and text book of hematology.3rded.Dr.Vipin Gupta/Avichal Publishing Company.Delhi:2014
21. Bhuyan A, Hazarika P, Deka R. Clinicopathological features of pancytopenia in adults and the role of bone marrow study in etiological categorization: A one-year cross-sectional study. Medical Journal of Babylon. 2022 Jul 1;19(3):415.
22. Halder B. Study of bone marrow examination findings in pancytopenia. [cited 2022Dec16].
23. Singh B, Joshi U, Kaushik A. Correlation of Bone Marrow Aspiration, Trephine Biopsy, and Imprint Cytology in Pancytopenia/Bicytopenia Cases in Resource-Limited Area. INTERNATIONAL JOURNAL OF SCIENTIFIC STUDY. 2021 Oct 1;9(4):139-43.
24. Ramanakumari Pasam, et al. A clinicohematological study of pancytopenia; IOSR Journal of dental and medical sciences; volume 15;issue 5 Ver IX(May 2016):71-76.
25. Taori, G., Ukey, A., & Bajaj, P. (2019). Comparison of Bone Marrow Aspiration Cytology, Touch Imprint Cytology and Bone Marrow Biopsy for Bone Marrow Evaluation at a Tertiary Health Care Institute. MVP Journal of Medical Sciences, 6(2), 152–157.
26. Javalgi AP, Dombale VD. Clinico – Hematological Analysis of Pancytopenia : A Bone Marrow Study. National Journal of Laboratory Medicine. 2013;2(4):12–7.

27. Santra G, Das BK. A cross sectional study of the clinical profile and aetiological spectrum of pancytopenia in a tertiary care center. Singapore Med J2010;51(10):806- 812.
28. Tejeswini V, Premalatha P, Renuka I V. Clinicohaematological profile of Pancytopenia- A South Indian tertiary hospital experience. Indian Journal of Pathology and Oncology. 2015;2(September):165–9.
29. Chandra S, Chandra H. Comparison of bone marrow aspirate cytology, touch imprint cytology and trephine biopsy for bone marrow evaluation. Hematol Rep 2011;3:65-7.
30. Pant S, Dhingra H, Gupta A, Misra RK. Comparative analysis of bone marrow aspiration smears, touch imprints and trephine biopsy in haematological malignancies. International Journal of Contemporary Medical Research [IJCMR]. 2020;7(6).

# ANNEXURES – I

**Ethical clearance**

IEC/no-09/2021
Date-22/01/2021

B.L.D.E. (DEEMED TO BE UNIVERSITY)
(Declared vide notification No. F.9-37/2007-U.3 (A) Dated 29-2-2008 of the MHRD, Government of India under Section 3 of the UGC Act, 1956)
The Constituent College
SHRI. B. M. PATIL MEDICAL COLLEGE, HOSPITAL AND RESEARCH CENTRE

## INSTITUTIONAL ETHICAL CLEARANCE CERTIFICATE

The Institutional ethical committee of this college met on 11-01-2021 at 11-00 am to scrutinize the synopsis of Postgraduate students of this college from Ethical Clearance point of view. After scrutiny the following original/corrected and revised version synopsis of the Thesis has been accorded Ethical Clearance

**Title:** A comparative study of bone marrow aspiration, imprints and bone marrow biopsy in pancytopenia

**Name of PG student:** Dr Anin Prakash, Department of Pathology

**Name of Guide/Co-investigator:** Dr R M Potekar Professor Professor of Pathology

DR .S.V.PATIL
CHAIRMAN, IEC
Institutional Ethical Committee
B L D E (Deemed to be University)
Shri B.M. Patil Medical College,
VIJAYAPUR-586103 (Karnataka)

**Following documents were placed before Ethical Committee for Scrutinization:**

1. Copy of Synopsis / Research project
2. Copy of informed consent form
3. Any other relevant documents.

Award No : 23
Date : 17/6/2021

# BLDE
## (DEEMED TO BE UNIVERSITY)
Declared as Deemed-to-be-University u/s 3 of UGC Act, 1956

The Constituent College

SHRI. B. M. PATIL MEDICAL COLLEGE, HOSPITAL & RESEARCH CENTRE, VIJAYAPURA

BLDE(DU)/REG/PG-Guide/2021-22/518 June 16, 2021

To,
The Professor and HOD
Department of Pathology,
BLDE (DU)'s Shri B. M. Patil Medical College,
Hospital and Research Centre,
Vijayapura

Sir,

Sub: Regarding change of PG Guide.
Ref: Your letter no. Path/2021/420 dated 1st June, 2021.

With reference to the subject and letter cited above, on approval of the Hon'ble Vice-Chancellor, the change of PG Guide is permitted in respect of PG Student of your department as per below:

| Sl. No. | Name of the Student | Previous Guide | New Guide | Batch/ Year |
|---|---|---|---|---|
| 1. | Dr. Sahitya H. | Dr. R. M. Potekar | Dr. S. U. Arakeri | 2018 - 19 |
| 2. | Dr. Saswati Subhadarshini | | Dr. Vijayalaxmi Patil | 2019 - 20 |
| 3. | Dr. Sultana Shahnaz | | Dr. S. B. Hipparagi | 2020 - 21 |
| 4. | Dr. Anin Prakash | | Dr. Savitri Nerune | 2020 - 21 |

This is for your information and needful.

REGISTRAR
REGISTRAR
BLDE (Deemed to be University)
Vijayapura-586103. Karnataka

Copy to:
- The Dean, Faculty of Medicine and Principal
- The Controller of Examinations
- The Concerned PG Teacher

Circulate to concerned PG guides & teachers.

Prof. & HOD
Dept. of Pathology
BLDE (Deemed to be University)
Shri B. M. Patil Medical College,
VIJAYAPUR-

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College: Phone: +918352-262770, Fax: +918352-263019, Website: www.bldedu.ac.in

## Annexure – II

## RESEARCH INFORMED CONSENT FORM

**TITLE OF THE PROJECT** : **A Comparative Study Of Bone Marrow Aspiration, Imprint And Biopsy In Pancytopenia**

**PRINCIPAL INVESTIGATOR** : Dr. ANIN PRAKASH

P.G. DEPARTMENT OF PATHOLOGY

**P.G.GUIDE** : DR.SAVITRI M. NERUNE(DCP, DNB),

ASSOCIATE PROFESSOR,

DEPARTMENT OF PATHOLOGY

**PURPOSE OF RESEARCH:**

I have been carrying out this study to assess the efficiency of bone marrow aspiration, imprint smears and biopsy in the diagnosis of pancytopenia cases.

**PROCEDURE:**

I understand that I will undergo detailed clinical history, thorough clinical examination and after which bone marrow examination will be performed to access the efficiency in diagnosing various causes of pancytopenia.

**RISK AND DISCOMFORTS:**

I understand that, I may experience pain and discomforts during the bone marrow examination. This is mainly the result of my condition and procedures of this study are not expected to exaggerate these feelings which are associated with usual course of treatment.

**BENEFITS:**

I understand that my participation in the study will have no direct benefit to me other than the potential benefit of the treatment.

**CONFIDENTIALITY:**

I understand that the medical information produced by the study will become a part of hospital record and will be subjected to confidentiality and privacy regulations of the hospital. If the data is used for publications the identity of patient will not be revealed.

**REQUST FOR MORE INFORMATION:**

I understand that I may ask more questions about the study at any time.

**REFUSAL FOR WITHDRAWAL OF PARTICIPATION:**

I understand that my participation is voluntary and that I may refuse to participate or may withdraw from the study at any time.

**INJURY STATEMENT:**

I understand that in the unlikely event of injury to me during the study I will get medical treatment but no further compensations. I have read and fully understood this consent form. Therefore I agree to participate in the present study.

_____ _____

Investigator Date

_____ _____

Signature of Witness Date:

**STUDY SUBJECT CONSENT STATEMENT:**

I confirm that **Dr. ANIN PRAKASH** has explained to me the purpose of research, the study procedure, that I will undergo and the possible discomforts as well as benefits that I may experience in my own language. I have been explained all the above in detail in my own language and I understand the same. Therefore I agree to give consent to participate as a subject in this research project.

_____ _____

(Participant) Date

_____ _____

(Witness to signature) Date

## Annexure - III

**PROFORMA**

| | | | |
|---|---|---|---|
| NAME | : | OP/IP No. | : |
| AGE | : | | |
| SEX | : | D.O.A | : |
| RELIGION | : | D.O.D | : |
| OCCUPATION | : | | |
| RESIDENCE | : | | |

**Presenting Complaints** :

**Past history** :

**Personal history** :

**Family history** :

**Treatment history** :

**General physical examination:**

Pallor present/absent

Icterus present/absent

Clubbing present/absent

Lymphedenopathy present/absent

Edema present/absent

Built poor/average/well

VITALS: PR: RR:

BP: TEMPERATURE:
WEIGHT:

**SYSTEMIC EXAMINATION:**

Cardiovascular system

Respiratory system:

Per Abdomen:

Central nervous system:

Clinical Diagnosis:

**INVESTIGATIONS**:

Haematological investigations:
Biochemical Investigations:
Radiological Investigations:

**Proforma for bone marrow aspiration :**

Preparation : Satisfactory /Adequate or Inadequate

Cellularity : Normocellular/ Hypocellular / Hypercellular Erythropoiesis
: Normoblastic /Megaloblastic/ Micronormoblastic. Myelopoiesis
: Morphology/ Number/ Maturation.
M:E ratio : Normal / Reversed.

Lymphocytes : Morphology/ Number/ Morphology. Megakaryocytes :
Normal/ Hypolobulated/ Hyperlobulated. Plasma cells : Present/ Absent/ Number.
Abnormal cells : If any.

Iron Stores : Grade of iron stores according to Gales method.

Serum ferritin : in ng/l

Impression :

## KEY TO MASTER CHART

| | | |
|---|---|---|
| BMA | - | Bone marrow aspiration |
| BMI | - | Bone Marrow Imprint |
| BMB | - | Bone Marrow Biopsy |
| PS | - | Peripheral smear |
| NCHC | - | Normocytic Hypochromic smear |
| MCHC | - | Microcytic hypochromic |
| F/A/O | - | Features Are Of |
| EH | - | Erythroid Hyperplasia |
| RPSIS | - | Right Posterior Superior Iliac Spine |
| LPSIS | - | Left Posterior Superior Iliac Spine |
| hpf | - | high power field |
| ng/ml | - | nano grams/ml |
| AML | - | Acute Myeloid Leukemia |
| CML | - | Chronic myeloid Leukemia |
| ALL | - | Acute Lymphoblastic Leukemia |
| CLL | - | Chronic Lymphocytic Leukemia |
| ITP | - | Idiopathic Thrombocytopenic Purpura |
| MDS | - | Myelodysplastic Syndrome |
| Sat | - | Satisfactory |
| Hyp | - | Hypercellular |
| Hypo | - | Hypocellular |
| Dilu | - | Diluted |
| Norm | - | Normocellular |
| m | - | months |
| cell | - | Cellular |

# Master Chart

| SL. NO. | Name | Age | Sex | IP number | Site | Peripheral Smear | BMA | BMB | BMA Impression | Special Stain | BMI Impression | BMB Impression |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | Preethi Siddangouda Patil | 21 | Female | 96 | RPSIS | NCNC anemia | BMA01/19 | BMB01/19 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 2 | Lakkawwa Goudappa | 65 | Female | 6567 | RPSIS | Dimorphic anemia | BMA04/19 | BMB2/19 | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC MATURATION |
| 3 | Sidaray Kasappa | 60 | Male | 87658 | LPSIS | NCNC anemia | BMA08/19 | BMB3/19 | ACUTE LEUKEMIA | MPO & PAS | - | ACUTE LEUKEMIA |
| 4 | Soujanya Manappa | 16 | Female | 3459 | RPSIS | Macrocytic anemia | BMA11/19 | BMB6/19 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | G3 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 5 | Gurabasayya | 20 | Male | 2345 | RPSIS | Macrocytic anemia | BMA18/19 | BMB11/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 6 | Gourabai | 52 | Female | 57657 | LPSIS | Macrocytic anemia | BMA19/19 | BMB12/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 7 | Ravi C Patil | 34 | Male | 4660 | RPSIS | NCNC anemia | BMA23/19 | BMB07/19 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |

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|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 8 | Virupakashi Sharanappa | 25 | Male | 8776 | RPSIS | Dimorphic anemia | BMA24/19 | BMB14/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 9 | Mayakka Yankappa Pujari | 22 | Female | 7800 | LPSIS | NCNC anemia | BMA29/19 | BMB10/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G4 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 10 | Charitra Mahesh | 24 | Female | 7784 | LPSIS | Dimorphic anemia | BMA35/19 | BMB15/19 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 11 | Sivarudra Siddagonda Biradar | 54 | Male | 54676 | RPSIS | NCNC anemia | BMA37/19 | BMB17/19 | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | G2 | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA |
| 12 | Shivanand Gangappa Kolur | 18 | Male | 87657 | RPSIS | Macrocytic anemia | BMA41/19 | BMB18/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 13 | ravi Gurappa | 23 | Male | 11694 | RPSIS | MCHC anemia | BMA43/19 | BMB20/19 | HYPOCELLULAR MARROW | - | APLASTIC ANEMIA | APLASTIC ANEMIA |
| 14 | Meenakshi Sangappa | 18 | Female | 9789 | STERNUM | Dimorphic anemia | BMA45/19 | BMB21/19 | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC MATURATION |
| 15 | Akshata Mallikarjun | 23 | Female | 1297 | LPSIS | NCNC anemia | BMA48/19 | BMB22/19 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | G5 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 16 | Kirshna Reven Siiddappa | 56 | Female | 15641 | RPSIS | Macrocytic anemia | BMA52/19 | BMB26/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G3 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |

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|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 17 | Hafisa Husenab Indikar | 70 | Female | 16545 | LPSIS | Macrocytic anemia | BMA57/19 | BMB29/19 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | G0 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 18 | Shivappa Sabu | 42 | Male | 116953 | RPSIS | Macrocytic anemia | BMA58/19 | BMB30/19 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G1 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 19 | Kantappa Nimbal | 44 | Male | 475811 | LPSIS | Macrocytic anemia | BMA01/20 | BMB01/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G4 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 20 | Vidya Naveen | 21 | Female | 42826 | RPSIS | Dimorphic anemia | BMA04/20 | BMB03/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 21 | Vishal Mohanan | 23 | Male | 1903 | RPSIS | NCNC anemia | BMA06/20 | BMB06/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G3 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 22 | Tipamma | 34 | Female | 4158 | RPSIS | MCHC anemia | BMA09/20 | BMB09/20 | ERYTHROID HYPERPLASIA WITH NORMOBLASTIC AND MICRONORMOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION |
| 23 | Mahesh Siddappa | 30 | Male | 4532 | LPSIS | Dimorphic anemia | BMA10/20 | BMB10/20 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | G3 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 24 | Parashuram Somaning | 30 | Male | 7529 | LPSIS | Macrocytic anemia | BMA14/20 | BMB14/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 25 | Mallappa | 78 | Male | 9872 | LPSIS | NCNC anemia | BMA20/20 | BMB16/20 | HYPOCELLULAR MARROW | G1 | HYPOCELLULAR MARROW | HYPOCELLULAR MARROW |

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|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 26 | Sabu Hannamat | 60 | Male | 10060 | RPSIS | Dimorphic anemia | BMA19/20 | BMB17/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 27 | Siriha Raju | 8 | Female | 12948 | RPSIS | Dimorphic anemia | BMA25/20 | BMB15/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 28 | Manalingappa | 85 | Male | 13239 | LPSIS | NCNC anemia | BMA27/20 | BMB13/20 | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | - | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA |
| 29 | Somanath Shankreppa | 30 | Male | 12992 | RPSIS | Dimorphic anemia | BMA26/20 | BMB18/20 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | G1 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION |
| 30 | Prashant Devindra | 27 | Male | 14557 | LPSIS | Dimorphic anemia | BMA29/20 | BMB19/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G4 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 31 | Vaibhav Vivek Dharane | 32 | Male | 14714 | LPSIS | Macrocytic anemia | BMA30/20 | BMB21/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 32 | ramesh | 12 | Male | 14807 | RPSIS | NCNC anemia | BMA31/20 | BMB20/20 | HYPOCELLULAR MARROW | G1 | HYPOCELLULAR MARROW | HYPOCELLULAR MARROW |
| 33 | Siddanna | 48 | Male | 17065 | LPSIS | Macrocytic anemia | BMA32/20 | BMB22/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 34 | Hanumanth | 66 | Male | 8850 | LPSIS | Dimorphic anemia | BMA35/20 | BMB23/20 | MYELODYSPLASTIC SYNDROME(MDS) | - | MYELODYSPLASTIC SYNDROME(MDS) | MYELODYSPLASTIC SYNDROME(MDS) |
| 35 | Vinod Kumar | 22 | Male | 14844 | RPSIS | Dimorphic anemia | BMA36/20 | BMB24/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |

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| 36 | Yallawwa | 45 | Female | 14829 | RPSIS | Dimorphic anemia | BMA37/20 | BMB25/20 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G0 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 37 | Nibisab | 36 | Male | 27481 | LPSIS | Dimorphic anemia | BMA39/20 | BMB25/20 | MYELODYSPLASTIC SYNDROME(MDS) | G2 | MYELODYSPLASTIC SYNDROME(MDS) | MYELODYSPLASTIC SYNDROME(MDS) |
| 38 | Abesha | 4 | Female | 69715 | LPSIS | NCNC anemia | BMA1/21 | BMB2/22 | ACUTE LEUKEMIA | - | ACUTE LEUKEMIA | ACUTE LEUKEMIA |
| 39 | Aishwarya | 10 | Female | 137764 | RPSIS | Dimorphic anemia | BMA7/21 | BMB7/21 | Dry tap | G4 | APLASTIC ANEMIA | APLASTIC ANEMIA |
| 40 | Mudakappa | 58 | Male | 72928 | LPSIS | Macrocytic anemia | BMA9/21 | BMB9/21 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G3 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 41 | Shridevi | 4 | Female | 72928 | RPSIS | NCNC anemia | BMA11/21 | BMB11/21 | HYPOCELLULAR MARROW | - | HYPOCELLULAR MARROW | HYPOCELLULAR MARROW |
| 42 | Rudragouda | 40 | Male | 78148 | LPSIS | Dimorphic anemia | BMA13/21 | BMB12/21 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 43 | Mallikarjun | 20 | Male | 106236 | RPSIS | Macrocytic anemia | BMA15/21 | BMB13/21 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G4 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 44 | Vijayakumar | 43 | Male | 120673 | LPSIS | Macrocytic anemia | BMA16/21 | BMB14/21 | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | G3 | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA | PLASMA CELL NEOPLASM S/O MULTIPLE MYELOMA |
| 45 | Prabhu B Sam | 30 | Male | 161338 | RPSIS | NCNC anemia | | BMB16/21 | Dry tap | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |

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| 46 | Sangeeta Manjunath Pujari | 3 | Female | 179530 | Tibia | MCHC anemia | BMA20/22 | BMB18/21 | HYPOCELLULAR MARROW | - | HYPOCELLULAR MARROW | HYPOCELLULAR MARROW |
| 47 | Mahadevi | 32 | Female | 180138 | RPSIS | Dimorphic anemia | BMA22/21 | BMB19/21 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 48 | Kusha Sharanappa | 18 | Male | 198590 | LPSIS | NCNC anemia | BMA24/21 | BMB20/21 | ACUTE LEUKEMIA | - | ACUTE LEUKEMIA | ACUTE LEUKEMIA |
| 49 | Suraksha Y | 9 | Female | 68356 | LPSIS | MCHC anemia | BMA1/22 | BMB21/21 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION |
| 50 | Ambika | 39 | Female | 75096 | LPSIS | NCNC anemia | BMA2/22 | BMB1/22 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | - | NORMOCELLULAR MARROW WITH MILD ERYTHROID HYPERPLASIA | NORMOCELLULAR MARROW WITH MILD ERYTHROID HYPERPLASIA |
| 51 | Rudramma | 29 | Female | 77210 | RPSIS | Macrocytic anemia | BMA3/22 | BMB2/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G1 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 52 | Mallapa | 50 | Male | 97003 | LPSIS | MCHC anemia | BMA4/22 | BMB3/22 | Dry tap | - | Filariasis | MYELOFIBROSIS |
| 53 | Shiva Gouda | 65 | Male | 101968 | RPSIS | MCHC anemia | BMA5/22 | BMB04/22 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | G2 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION |
| 54 | Gouramma | 52 | Female | 15049 | LPSIS | NCNC anemia | BMA6/22 | BMB5/22 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC AND MEGALOBLASTIC MATURATION |
| 55 | Sharndasu | 17 | Male | 145741 | RPSIS | Dimorphic anemia | BMA8/22 | BMB8/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |

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| 56 | Devendra Choudari | 28 | Male | 162872 | LPSIS | NCNC anemia | BMA9/22 | BMB6/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 57 | Laxmi | 11 | Female | 167113 | LPSIS | MCHC anemia | BMA10/22 | BMB07/22 | Dry tap | G1 | APLASTIC ANEMIA | APLASTIC ANEMIA |
| 58 | Anand | 22 | Male | 192019 | RPSIS | Macrocytic anemia | BMA12/22 | BMB9/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 59 | Bhagyashree | 18 | Female | 23600 | LPSIS | Dimorphic anemia | BMA17/22 | BMB13/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | G1 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 60 | Laxmi | 14 | Female | 216640 | LPSIS | Dimorphic anemia | BMA20/22 | BMB15/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 61 | Umash | 45 | Male | 238215 | RPSIS | Macrocytic anemia | BMA22/22 | BMB17/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 62 | Chandrakala | 7 | Female | 246626 | RPSIS | NCNC anemia | BMA23/22 | BMB18/22 | ACUTE LEUKEMIA | G1 | ACUTE LEUKEMIA | ACUTE LEUKEMIA |
| 63 | Laxman | 23 | Male | 255542 | RPSIS | NCNC anemia | BMA24/22 | BMB19/22 | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MEGALOBLASTIC MATURATION |
| 64 | Mallikarjun | 35 | Male | 291955 | RPSIS | MCHC anemia | BMA25/22 | BMB20/22 | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | - | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION | ERYTHROID HYPERPLASIA WITH MICRONORMOBLASTIC MATURATION |

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